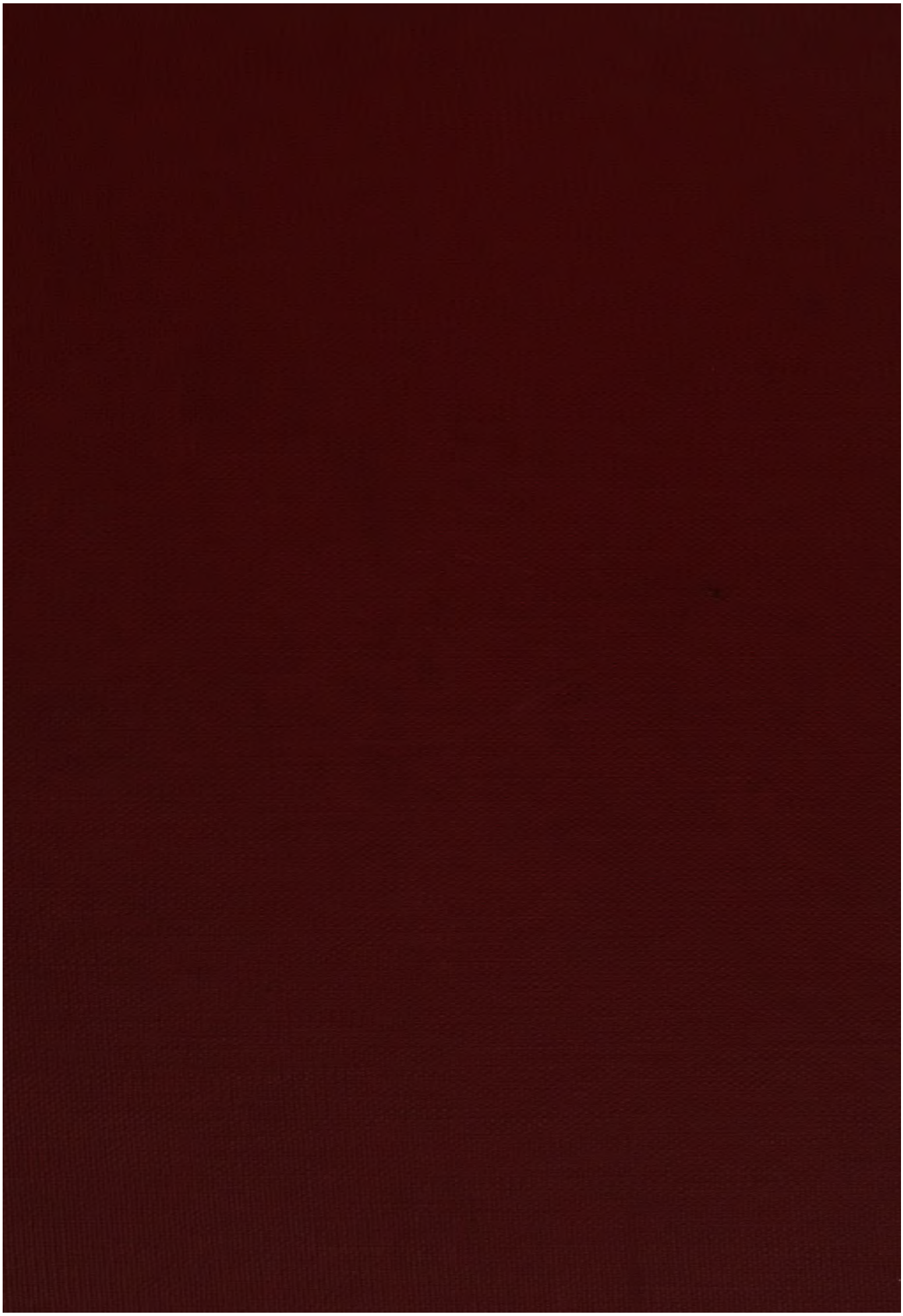


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THE UNIVERSITY OF LIVERPOOL

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ISSUED BY THE

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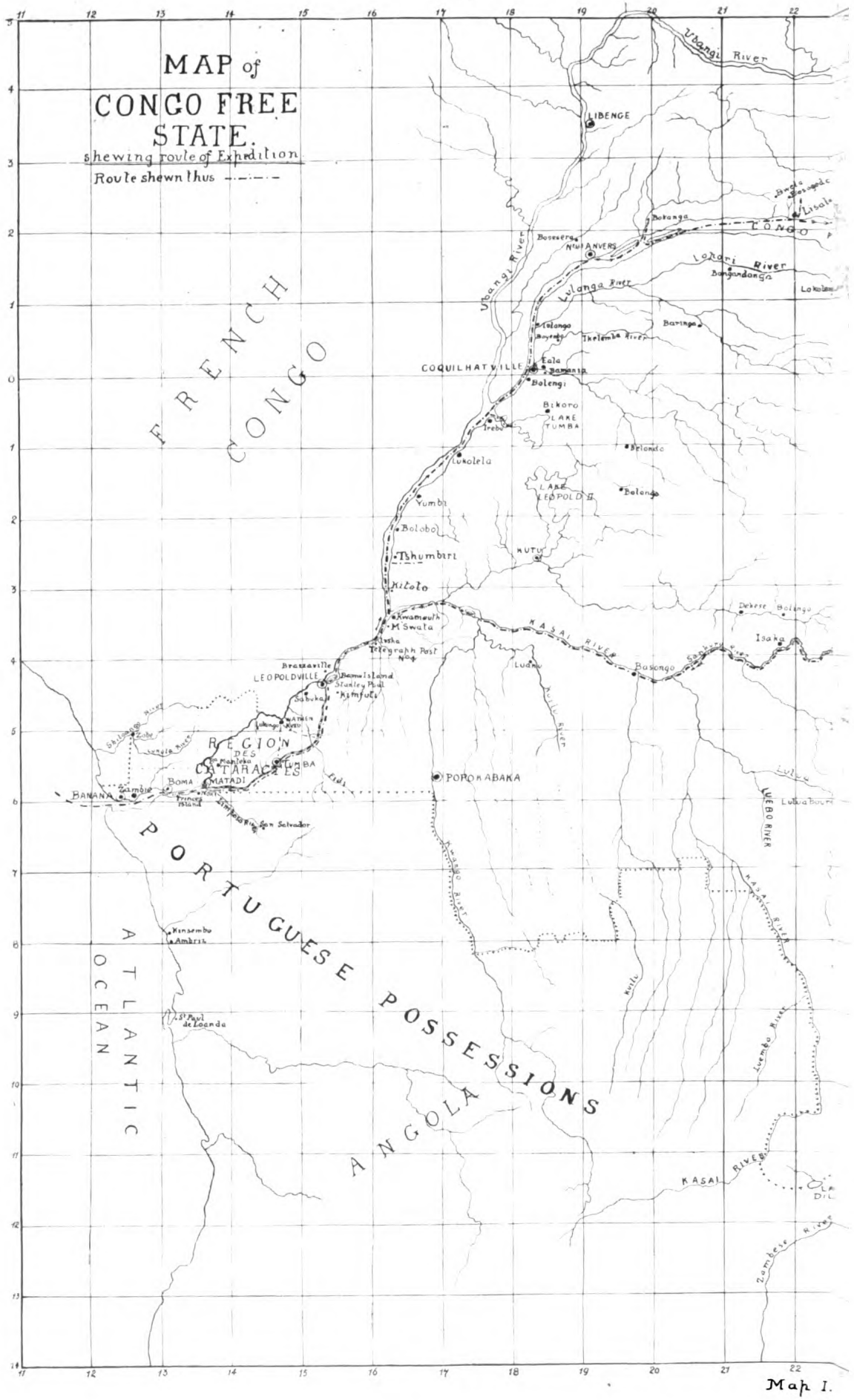
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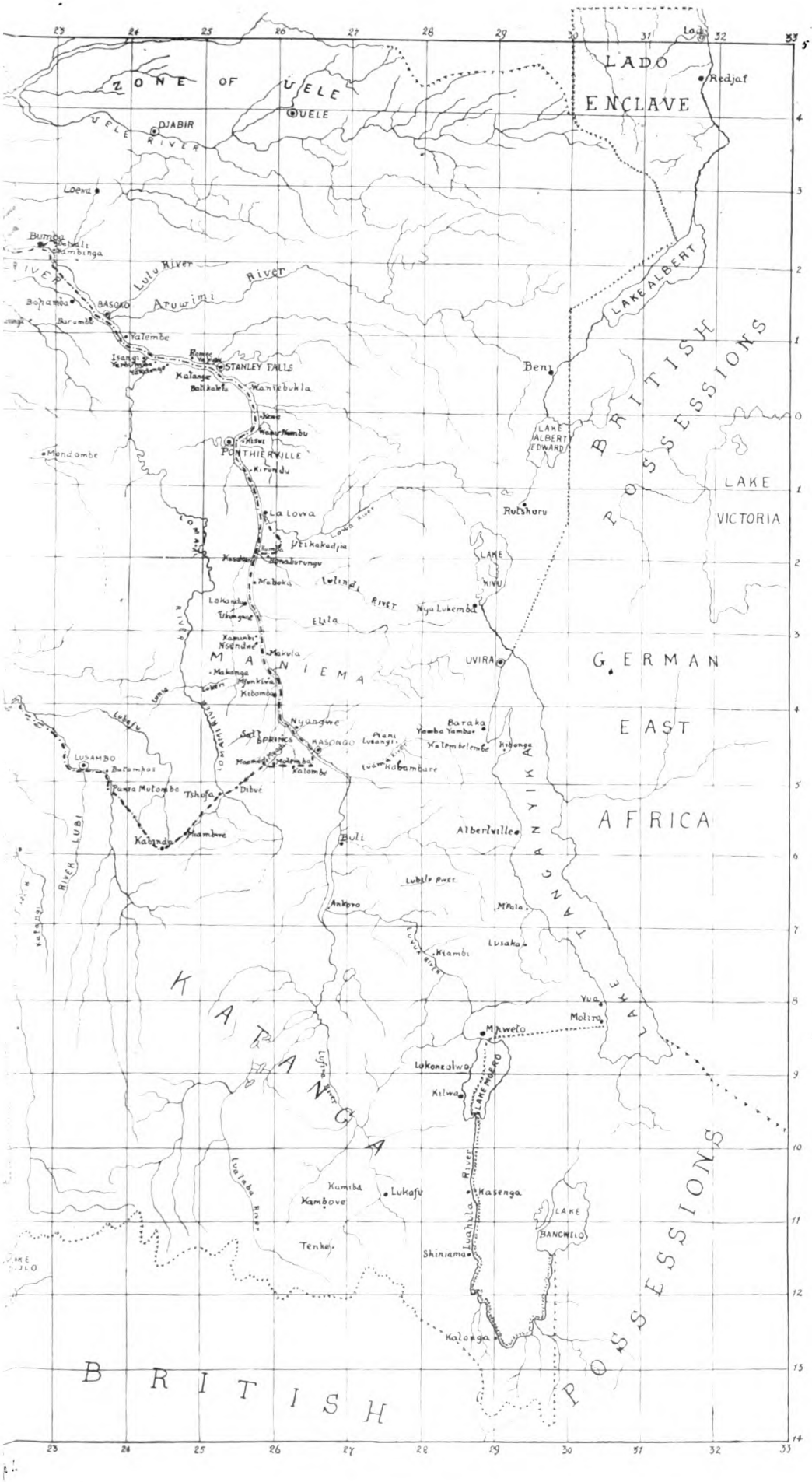
By order of the Committee of the Incorporated Liverpool School of Tropical Medicine, the series of the Reports of the School, which have been issued since 1899, will be followed, from January 1, 1907, by the Annals of Tropical Medicine and Parasitology, of which this is the first number.

The Annals are issued by the Committee of the School, and will contain all such matter as was formerly printed in the Reports—that is to say, accounts of the various expeditions of the School and of the scientific work done in its laboratories at the University of Liverpool and at Runcorn. Altogether twenty-one Memoirs, besides other works, have been published by the School since 1899, and of these ten, containing 519 quarto or octavo pages and 95 plates and figures, were published during the two years 1904 and 1905; and there is no reason to suppose that this rate of production by the workers of the School alone will diminish in the future. In addition, however, to School work, original articles from outside on any subject connected with Tropical Medicine or Hygiene may be published if found suitable (see notice on back of cover); so that, in all probability, not less than four numbers of the Annals will be issued annually. Each number will be brought out when material sufficient for it has been accumulated.

INSECTS AND OTHER ARTHROPODA COLLECTED IN THE CONGO FREE STATE


MAP of
CONGO FREE STATE.
shewing route of Expedition.
Route shewn thus
FRENCH CONGO
PORTUGUESE POSSESSIONS
ANGOLA
ATLANTIC OCEAN
Ubangi River
LIBENGE
Bosesera
ANVERS
Bokanga
LONGO
Lisala
Lokari River
Bangandanga
Lokolela
Lulanga River
Ubangi River
Baringa
Ikelemba River
COQUILHATVILLE
Eala
Bamania
Bolengi
Bikoro
LAKE TUMBA
Irebu
Lukolela
Belonde
LAKE LEOPOLD II
Bolenge
Yumbi
Bolobo
Tshumbiri
KUTU
Kitoto
Kwamouth
M'Swata
Isoko
Telegraph Post No 4
Dekese
Bolenge
KASAI RIVER
Basongo
Isaka
Brazzaville
LEOPOLDVILLE
Barrows Island
Stanley Pool
Kimfuta
Sabuka
Ludima
Lulua River
Lubefu River
Zaire River
Zaire
Lukaya River
REGION DES CATARACTES
Kinchassa
Lukunga
MATADI
Lubefu
Lulua
Lulua Bourg
LUEBO RIVER
BOMA
Nokki
Princes Island
BANANA
Kwilu
Kongo River
San Salvador
POROMABAKA
Kinsembo
Ambrizette
St Paul de Loanda
Kasai River
Kwilu
Luembe River
Zambese River

Map I.


ZONE OF UELE
LADO ENCLAVE
Redjaf
UELE
DJABIR
UELE RIVER
Loeni
Lulu River
Aruwimi River
BASOKO
Bafwabogo
Yalombe
STANLEY FALLS
Kalanga
Wanikebukla
Mondombe
PONTHIERVILLE
Kisui
Kirundu
La Lowa
Ikelemba
Maboko
Lulindi RIVER
Nya Lukemba
Elila
Maxwia
MANIEMA
Makinga
Kibombo
Nyangwe
KASONGO
Baraka
Yamba Yambo
Kalembelembe
Lusambo
Batempas
Tshofa
Dibue
Kabanda
Buli
Albertville
Lubilo River
Ankoro
Mpala
Lusaka
Kiambi
KATANGA
Yua
Moliro
Mpweto
Lukonzolwa
Kilwa
LAKE MOERO
Kamiba
Kambove
Lukafu
Kasenga
Tenke
Shiniama
LAKE BANGWELO
Kalonga
BRITISH POSSESSIONS
LAKE ALBERT
LAKE ALBERT EDWARD
Beni
Rutshuru
LAKE KIVU
UVIRA
GERMAN EAST AFRICA
LAKE VICTORIA
RIVER LUBI
Lualaba River
Luapula River
Luvubhla River

INSECTS AND OTHER ARTHROPODA COLLECTED IN THE CONGO FREE STATE

Being the Seventh Interim Report of the Expedition of the Liverpool School of Tropical Medicine to the Congo, 1903-05

BY

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(WALTER MYERS FELLOW, UNIVERSITY OF LIVERPOOL)

AND

JOHN L. TODD, B.A., M.D., C.M. McGILL
(DIRECTOR OF THE RUNCORN RESEARCH LABORATORIES OF THE LIVERPOOL SCHOOL OF TROPICAL MEDICINE)

INTRODUCTORY

This expedition was sent to the Congo Free State, on the invitation of His Majesty King Leopold II, by the Liverpool School of Tropical Medicine to report upon the sanitary condition of the more important posts and to study human trypanosomiasis. The collection of various biting insects formed, therefore, a principal part of its work; but once the more important ones, as *Glossina* and *Anophelina*, were obtained in each district the systematic search for insects was discontinued. A large part of this collection represents then, merely the casual wayside gleanings made during a journey through Central Africa.

From the point of view of a dipterologist the collection is therefore regrettably incomplete, especially so, since a large number of specimens have been unavoidably ruined by moulds. Had time been spent in collecting, it is probable that many of those insects reported from only one or two localities would have been found to have a far wider distribution.

Twenty-three months were actually spent in the Congo Free State; during that time a distance of some 2,000 miles was travelled. The course followed by the expedition is indicated on the map; while

the dates when it was at, or passed through, the various posts are briefly mentioned below.

England was left on September 4, 1903. Banana,* the first point of call in the Free State, was reached on September 23. From September 24 to October 27 was spent at Boma, and from October 28 to November 20 at Matadi. Matadi is connected with Leopoldville by railroad. From Tumba, the half way point on the railway line, two of the members of the expedition made a trip of three weeks' duration (November 5 to 28) to Wathen and through the District of the Cataracts. A stay of seven months, from November 22, 1903, to June 23, 1904, was made at Leopoldville.

From Leopoldville the expedition went up the Congo to Stanley Falls in a small steamer placed at their disposal by the Government. When it seemed advisable, stops and short side-journeys were made at posts along the river for the purpose of studying local conditions. Work was done at M'Swata from June 25 to 29. At Tshumbiri (July 2 to 5) one night was spent at a native village some ten miles inland. Mopolengue was visited on July 7 to 9, and Irebu on July 14 to 17. From Coquilhatville (July 18 to August 9) trips were made to Eala and Bamamia. Nouvelle Anvers was visited from August 14 to 21. From Mobeka (August 22) a short journey was made up the Mongala to Bokanga (August 23). Stops were made at Lisala (August 26 to 30) and at Bumba (August 31 to September 4); at both of these places short journeys were made inland. Short stays were made at Basoko (September 7 to 9) and at Yakusu (September 13 to 15). On September 15 Stanley Falls was reached, where the expedition remained until September 28.

From Stanley Falls the journey up the Congo was continued to Kasongo by canoe. Ponthierville was visited from October 5 to October 9. From Lalowa (October 12) two members of the expedition made a side trip of 12 days' duration inland to the east of the Congo. They paralleled the river as far as Kumba, where they once more took canoe. From Lokandu (October 16 to November 1) a journey of some 18 miles was made inland. Stops were made at N'Sendwe (November 4 to 8), and Nyangwe (November 13 to 22). Kasongo was reached on November 23, 1904. A stay of five months

* See Map 1, page 1.

was made at this post, and it is there that Dr. Dutton died and is buried.

Kasongo was left on April 27, 1905. The journey thence to Pania Mutombo lay overland and was done on foot. Kalombe was reached on May 3, Maomedi on May 5. No stop of more than a day was made before reaching Tshofa (May 13 to 25). Four days, June 2 to 6, were passed at Cabinda. At Pania Mutombo (June 12) canoes were once more employed for the two days' journey down stream to Lusambo (June 5), where the expedition remained until July 8, 1905.

The journey from Lusambo to Leopoldville was done as quickly as possible by steamer, and no stops were made for work. At Leopoldville (July 21 to 30) arrangements were made for leaving Africa. The railroad between Leopoldville and Matadi was once more traversed, and on August 8, 1905, the expedition left the Congo Free State. On the way home Freetown, Sierra Leone, was visited for a week (August 16 to 22). Liverpool was reached on September 5, 1905; the expedition had been absent from England for just two years.

Other side-trips were made besides those mentioned; and it must be understood that when a stay of any duration was made in a post short journeys were always made in its neighbourhood.

As is shown by the map, the route followed by the expedition twice crossed the equator and traversed a territory lying between 2° North and 6° South of that line. The climates of the posts visited therefore varied somewhat. The succession of the seasons at the most important places is briefly as follows:—

At Boma the seasons are irregular. There is usually said to be a long and a short wet, and a long and a short dry, season. Practically, we may say that from May to September there is but very little—or no—rain; and that from October to the end of April there is a good deal; from January to April are the hottest months of the year. It is then, from October to May that conditions are the most favourable for the breeding of mosquitoes. At Leopoldville rain is more likely to fall in every month of the year than at Boma. From the 15th of June to the 1st of September is usually considered to be the dry season, and during this period there may be practically no rain.

Coquilhatville is almost on the equator, and consequently has no marked variation in its seasons. Rain falls in every month of the year, perhaps the most in November and December. The

temperature is constantly high, and during the first four months of the year it may be very hot. The climate is then admirably suited for the development of mosquitoes.

Bumba is one of the furthest north among the posts visited by the expedition. The seasons there are not well marked, rain falls in every month of the year; perhaps the least in January and February.

At Kasongo and Cabinda the seasons are much the same as at Boma, save that the "dry season" is shorter and rain often falls in August and May.*

When the expedition arrived at Boma the rainy season had already set in. For the first half of the month spent there, rain fell almost daily, the thermometer meanwhile varying between 85° and 70°F. During the latter two weeks there was an occasional shower or two and the temperature was slightly higher.

While at Matadi there was but little rain, so that but few of those places were seen where rain-water collects during the wet season to form breeding-places for mosquitoes.

When Leopoldville was reached the rainy season was well-established. The ground was sodden with water and the streams were filled to overflowing. From November to April rain fell almost daily, and there were frequent thunderstorms. The daily temperature varied from 70° to 89°F. In May less rain fell, and in June the "dry season," with its accompanying over-cast sky, evidently commenced, although there were still occasional thunderstorms.

On leaving Leopoldville practically no rain was met until the neighbourhood of Coquilhatville was reached. While the expedition was there rain fell almost every second day. One or two really hot days, maximum temperature 93° to 94°F, were encountered on the way up river. At Coquilhatville the temperature was much lower and exceedingly equable; this type of weather persisted during the journey up river, past Bumba and Basoko, to Stanley Falls. There

*For general information concerning the climate of the Congo Free State we are indebted to a "Rapport sur le Climat, la constitution du sol et l'Hygiène de l'Etat Indépendant du Congo" presented to the "Congrès national d'Hygiène et de Climatologie" at Brussels, Aug. 14, 1897, by Drs. Bourguignon, J. Cornet, G. Dryepondt, Ch. Firket, A. Lancaster and E. Meuleman. Thanks are also due to Dr. Etienne for copies of his meterological observations at Banana, and to Commandant Verdick for a copy of the observations made during 1904-05 at Kasongo.

were, however, one or two hot days (maximum 96°F), and while at the Falls there were a couple of severe thunderstorms.

From Stanley Falls the route lay south through the equatorial zone towards Kasongo where the rainy season had already set in. Frequent showers and thunderstorms were encountered on the way up river. The weather was pleasantly cool, the temperature varying usually between 82° and 71°F.

From November to April there was much rain at Kasongo (December, 216·3 mm.). The temperature meanwhile varied between 68° and 85°F. The march overland from Kasongo to Lusambo in May and June was made during the commencement of the dry season. Rain fell but rarely, the temperature became higher and its daily variation greater. Streams and swamps dried up rapidly, and by the end of the first week in May the prairie grass was often dry enough to burn.

Rain fell only twice during the stay of the expedition at Lusambo.

By referring to this synopsis of the weather encountered by the expedition the climatic conditions under which the insects, described in this report, were collected may be easily ascertained.

In the case of a post at which a long stay was made the month in which the specimen was obtained has been specified. To facilitate the finding of places in the map, the names of localities at which specimens were collected are always mentioned in the order in which they were visited.

A certain number of insects were sent to the expedition from places it left unvisited. In referring to these specimens the name of the collector and, when possible, the date of capture are given.

A considerable number of flies, collected in the British Colony of the Gambia* and in French Senegal, were taken, together with certain drawings and observations referring to them, to the British Museum on the return of the expedition of the Liverpool School of Tropical Medicine to Senegambia in July, 1903.

At the end of 1905 these insects had not been described by the British Museum authorities. It was therefore requested that they should be returned to the Liverpool School. While at the Museum, a considerable part of the collection had unfortunately been mislaid;

*For a map showing the localities mentioned in the Gambia, see Memoir XI of the Liverpool School of Tropical Medicine.

but a description of the remaining insects, which were returned to Liverpool, has been incorporated in this publication, with one or two exceptions, and these will be subsequently referred to as occasion may arise.

We desire to express our indebtedness and thanks to Mr. F. V. Theobald and Mr. E. E. Austen. for kind assistance rendered in the identification of the more obscure insects; and to Professor G. Neumann for the identification of the Ixodidae, and also for his valuable contribution to this group of the Arthropoda.

FAMILY CULICIDÆ

SUB-FAMILY ANOPHELINÆ

Pyretophorus costalis, Loew

Localities:—Zambie; Boma (Oct. 8 to 21); Prince's Island; Matadi (Oct. 29 to Nov. 25); Tumba; Wathen; Leopoldville (Dec. 10, 1903, to June 16, 1904); Telegraph post No. 4; Kitoto; Yumbie; above Lukolela in bush; Irebu; Coquilhatville; Bamamia; Lulongo; Nouvelle Anvers; Bokanga; Lisala; Bumba; Yambinga; below Basoko in bush; Stanley Falls; Benaburungu; Lokandu; Sendwe; Makula; Kasongo (Nov. 26, 1904, to Feb. 13, 1905); Tshofa; Lusambo; Lado enclave (Nov. Lemaire).

Circumstances of capture:—Imagines were taken in the day time and at night in the dwelling-places of Europeans and natives, in the mosquito nets of servants sleeping in the bush and, during the day time, far in the forest itself at a long distance from any village. Several were also caught in the evening about the table lamp on board our steamer.

Breeding places:—Pupae and larvae were found amongst the aquatic grasses growing along river edges, in swamps, in small clear puddles of rain-water lying on clayey soil, and in the foul-smelling pools used by the natives of the lower Congo for steeping manioc.

Field notes:—This is by far the most common Anopheline in the Congo. It seems to feed most fiercely just after sunset and again before dawn.

On two occasions during the stay of the expedition at Leopoldville it was necessary to work all night. On each occasion *P. costalis* was noticed to feed from dusk to about 11 p.m. From 11 p.m. to 3 a.m. none were seen. From 3 a.m. to

5 a.m. they fed fiercely, 15 or 20 insects attacked one at the same moment, and were able to feed successfully through clothing.

At Coquilhatville, early in the evening, these mosquitoes were seen flying into the houses of Europeans where ordinarily none could be found during the day time. The parasites of malaria were seen to develope in this mosquito at Boma.

In some of the mosquito breeding pools at Leopoldville the larvae of *P. costalis* were completely destroyed by larger, cannibalistic *Culex* larvae.

At Leopoldville in December, 1903, and in July, 1905, as well as at Ponthierville in October, 1904, it was observed that *Culicidae* were not breeding in the water of puddles and small streams containing an amorphous, brownish, apparently vegetable deposit,* although similar neighbouring collections of water contained many larvae.

Pyretophorus marshallii, Theobald

Six specimens, all females, were captured in the localities mentioned below. They were in all cases associated with *P. costalis*, but may readily be distinguished from the latter by the characteristic banding of the palpi. It is important to note, however, that among the long series of *P. costalis* there are many intervening forms between typical examples of the two species.

Localities:—Boma; Leopoldville (May); Coquilhatville; Yambinga.

Circumstances of capture:—The specimens were taken in the huts and mosquito nets of natives.

Myzomyia funesta, Giles

Localities:—Zambie; Prince's Island; Matadi; Wathen; Kalombe; Lusambo.

Circumstances of capture:—Examples of this mosquito were taken in the houses of both Europeans and Africans as well as in the mosquito nets of native servants. It is curious to note that not a single specimen of this mosquito was seen at Leopoldville where a careful mosquito survey was made.

Breeding places:—Examples of this species were obtained from larvae developed in the laboratory. It is probable that in the Lower Congo it breeds amongst aquatic grasses along the banks of rivers and in pools used for steeping manioc.

* Possibly a species of *Crenothrix* on whose gelatinous sheath iron becomes deposited as ferric oxide.

Field notes:—Malaria parasites were seen to develope in these mosquitoes at Lusambo.

Myzorhynchus paludis, Theobald

Localities:—Leopoldville (Dec. 1903 to Feb. 1904); Bamu Island; Bamamia; Eala; Barumbu; Kumba; Kasongo (Dec.); Lusambo.

Circumstances of capture:—Imagines were caught in forests and marshes and in natives' huts. They also came on board the steamer in the evening after the lamps were lighted, and at Lusambo were caught about the lamps on three occasions.

Breeding places:—Adults were bred from larvae taken in marshes and in the stagnant and overgrown but fairly clean water left in pits from which clay had been taken for brick-making.

Myzorhynchus mauritianus, Grandpré

Localities:—Zambie; Boma; Leopoldville (Dec.); Bamu Island; Kasongo (Dec.).

Circumstances of capture:—Imagines were caught only in the forest at Bamu Island.

Breeding places:—Larvae and pupae were taken from among the grass along the edges of rivers, from swamps, from rain-water collected in puddles on clayey ground, and from stagnant, overgrown, but fairly clean, water left in pits from which clay had been taken for brick-making, and imagines were bred from them.

Cellia pharoensis, Theobald

Localities:—This mosquito was collected only at Boma. Specimens were caught in dwelling-places and some were also reared from larvae.

Breeding places:—Larvae were found amongst water-grasses growing along the river's edge, in swamps and in a dirty, muddy puddle

Field notes:—Malaria parasites were seen to develope in this mosquito at Boma.

SUB-FAMILY TOXORHYNCHITINÆ, Theobald

Toxorhynchites marshallii, Theobald

FEMALE.—*Head* scales bronzy-blue at the sides and between the eyes, bronzy-green on occiput, black along the nape. *Palpi* of four segments; sub-apical and apical segments minute; clothed with brilliant azure blue scales. *Antennae* black; first segment black at the base dorsally, grey at the apex with a patch of dull white scales; pubescence grey; hairs black. *Proboscis* azure blue. *Thorax:* Prothoracic lobes dull azure blue, with bronzy-green reflections; mesothorax black with brilliant bronzy-green scales; scutellar scales rich bronzy-yellow; and there are patches of the same coloured scales near the base of the wings; pleurae almost covered with dull white scales. *Abdomen* azure blue with rich violet reflections; penultimate segment with brilliant ruby reflections; anterior half of anal tuft black, the rest rich orange; there are white lateral spots to the first, second, third and fifth segments; venter blue with white scales on the fifth segment. *Legs* dark violet; coxae with some white scales; mid and hind femora dull bronzy-yellow beneath with white reflections, in some lights, and in the hind tibiae this colour extends to the upper surface on the inside; first segment of mid and hind tarsi with a dusky white band; second segment almost entirely white forming a distinct broad band; second tarsal segment to fore legs with an inconspicuous basal band of dusky white scales; fore, mid and hind ungues equal and simple.

Length 9 to 10 mm., exclusive of the proboscis.

A description of the ♀ has been given as it has not, apparently, been hitherto described.* Of the 22 specimens collected, only one is a male, the rest are all females.

Localities:—Tshumbiri and Coquilhatville.

Circumstances of capture:—Our thanks are due to the Rev. and Mrs. Billington, members of the American Baptist Missionary Union stationed at Tshumbiri, for a series of these mosquitoes collected in March, April, June and November. They have also supplied the following notes on the bionomics of these mosquitoes. "The adults fly with a characteristic loud humming; they were frequently caught in a European's house. The larvae were found in a metal water-tank."

* Mono. Culicid., vol. 3, p. 121, 1903.

SUB-FAMILY CULICINÆ

Eretmapodites inornatus, n. sp. (Newstead)

(Pl. i, fig. 10)

Head clothed with brilliant silvery metallic scales with a patch of black ones in the centre. Thorax rich brown with two median, two short lateral, and a continuous marginal stripe of golden-yellow scales. Abdomen black; penultimate segment of the male with two lateral silvery spots; apical segment, in female only, almost entirely covered by silvery metallic scales; venter with five white metallic bands. Legs long, black with coppery or bronzy-brown reflections; no paddles to hind tarsi of ♂.

FEMALE.—Large basal median area of *head* clothed with flat black scales and a few isolated metallic silvery ones; upright forked scales black, rigid, occupying the dark area only; nape with a few narrow-curved, golden-yellow scales; lower third at the sides with flat black ones. *Palpi* densely clothed with black scales. *Proboscis* black with beautiful peacock-blue reflections. *Thorax:* Prothoracic lobes clothed with flat silvery scales except at base where they are black, there are also a few outstanding yellow scales and three long black bristles; mesothorax covered with dark brown and golden-yellow curved scales; the latter forming two median, two lateral and a continuous marginal stripe; the median stripes are short and curved downwards almost touching the marginal stripe; mid-lobe of scutellum with a long median patch of silvery metallic scales, bordered by narrow-curved, black ones; lateral lobes with a tuft or rosette of narrow-curved golden-yellow and a few black scales; metanotum with three black chaetae and three minute narrow-curved, golden-yellow scales (in type ♀); pleurae brownish-yellow or ochreous with a well-defined silvery line. *Abdomen* black, basal half bronzy-brown and black in some lights; penultimate segment with two lateral silvery spots; terminal segment silvery, tipped with black; venter black with five brilliant silvery bands and two lateral terminal silvery spots; the terminal band is interrupted in the centre by a narrow line of golden scales, which are continued into the succeeding segment where they form a rectangular basal patch. *Legs* black, bronzy-brown or peacock-green in some lights; tarsi paler; coxae ochreous; knee spots to posterior pair more conspicuous than in the anterior and mid pairs.

Wings clothed with dark brown scales, but in strong lights are of a beautifully bronzy peacock-blue and green.

Length 4·50 to 6·50 mm.

MALE. *Head* as in the ♀ but the narrow-curved golden scales are more numerous and extend over the whole of the black median patch. *Thorax:* Prothoracic lobes and mesothorax as in the ♀ ; scutellum rubbed in both specimens, but there are traces of a similar decoration to that which is seen in the ♀ ; metanotum nude.* *Abdomen* slender, cylindrical; black with peacock-blue reflections; antepenultimate segment enormously dilated, with two large, lateral, silvery patches. Genitalia with the clasps of great length, and the slender basal segment densely clothed with hairs and scales; second segment with a long apical spine.

Length about 4 mm.



FIG. 1. *Eretmapodites inornatus.* WING OF FEMALE. × 40.

Distinguished from *E. quinquevittatus*, Theob., by the presence of narrow-curved golden-yellow head scales, and the absence of paddles to the hind tarsi of the ♂ besides other important details. Theobald makes no reference to the presence of chaetae and scales on the metanotum of *E. quinquevittatus* either in his monograph or in the Genera Insectorum, but they are quite evident in this species and also in *E. austenii*,† Theob.; at the same time they are not traceable in the males of either of these species, their absence may, however, be due to abrasions.

* There is no trace of scales or chaetae in either of the two males which were collected.

† Mono. Culcidae, vol. 1, p. 283; this species awaits description.

Localities:—Coquilhatville; Lusambo.

Circumstances of capture:—Adults were caught only in the bush near water. On one occasion they were found in large numbers near a native village. They fed viciously at five in the afternoon; none of some 30 males and females caught at that hour appeared to have previously fed on blood.

Eretmapodites austenii, Theobald

Localities:—Coquilhatville; Stanley Falls; Kasongo.

Circumstances of capture:—Adults were twice caught in the afternoon in European houses. They were also taken in the "bush" near a native village placed on a small forest-covered island; they fed viciously at five in the afternoon.

Stegomyia fasciata, Fabricius

Localities: Matadi; Wathen; Leopoldville (Dec., 1903); Sendwe; Kasongo (April); Tshofa; Lusambo.

Circumstances of capture: Adults were caught during the day in the houses of Europeans.

Breeding places:—Adults were hatched in the laboratory from larvae and pupae taken from water collected in old pots and tins and from a foul-smelling pool used for steeping manioc.

Stegomyia argenteopunctata, Theobald

Localities:—Matadi; Kwamouth; Tshumbiri; Kisui.

Circumstances of capture:—Adults were caught only during the day time in the thick forest. At Kwamouth they were exceedingly numerous about a forest spring and bit viciously at mid-day. Those caught in a similar spot at about noon at Kisui did not attempt to feed. There were no huts within 250 yards of either of these places.

Breeding places:—A larva, allowed to develope in the laboratory, was taken from among the grasses along the river's edge at Matadi.

Stegomyia simpsoni, Theobald

Localities: Matadi; Wathen; Leopoldville (Dec.); Tshumbiri; Kasongo.

Circumstances of capture:—Adults were taken, while feeding, in the houses of Europeans in the afternoon and at night (7.30 p.m., lamps lighted).

Breeding places:—Larvae were taken from water collected in old pots and tins and from a font containing holy water in a mission building. Imagines were bred from them.

Stegomyia luteocephala, n. sp. (Newstead)

(Pl. ii, fig. 5)

Head yellow. Palpi black with white tips. Thorax brown, with two large, anterior, lateral silvery spots, a median yellow stripe, and posterior lateral yellow spots; scutellum white. Abdomen black with pale narrow bands, terminal segments silvery. Legs black with silvery spots and white-banded tarsi.

FEMALE.—*Head* with the large central area thickly clothed with large, loose, flat yellow scales, gradually merging into smoky-yellow in front; a narrow silvery-white line to the anterior half of the eyes formed of a single series of broad, flat, closely appressed scales; between the marginal line and the central yellow patch is a broad band of brownish-black scales, from which, anteriorly, arise several upright forked scales; nape with a few long, thick, straight or slightly curved, pale golden scales, on either side of which is a group of upright-forked scales, intermixed black and yellow; lower basal portion with flat, dusky-white scales, the marginal ones forming two dull silvery spots, sharply divided by a dense black spot. *Antennae* black, nodes white; hairs black; pubescence grey. *Palpi* black; tips with long silvery-white scales. *Thorax*: Prothoracic lobes with flat silvery-white scales; mesothorax with a well-defined median line and two lateral spots of narrow-curved, golden-yellow scales; anteriorly there are also two large spots of flat silvery-white scales, and a few silvery-white scales on the lower margin of the posterior yellow spots; the rest of the mesothorax with rich dark brown scales; scutellum with flat silvery-white scales; pleurae dark brown with two large patches of silvery scales. *Abdomen* rich bronzy-brown; segments 1 to 6 each with a well-defined, narrow, basal band of smoky-yellow scales; penultimate segment with a large lateral patch, and the terminal segment almost covered with brilliant metallic-silvery scales; venter with well-defined more or less triangular patches of metallic-silvery scales narrowing towards the apex, where they appear as two divergent lines; the scales forming the outer lateral angles of the spots projecting at the sides of the abdomen appearing *as outstanding scales*. *Legs* bronzy

blackish-brown; coxae and trochanters, ochreous; anterior and mid femora with scattered metallic-silvery scales; hind femora with a central anterior band and an apical group of silvery scales; anterior and mid tarsi with narrow dull-white basal bands to the first three segments, metatarsal band broadest; hind tarsi with a broad white basal band to the first, a narrow one to the second, and the third segment almost entirely white above, basally it is not so. *Wings* uniformly pale brown rather densely scaled, first sub-marginal cell much longer and slightly narrower than the second posterior.

Locality:—A single specimen of this species was caught in the bush at Kumba.

Stegomyia albomarginata, n. sp. (Newstead)

(Pl. ii, fig. 4)

Head black, anterior margin white. Thorax grey-brown; pleurae grey. Abdomen dark brown with lateral grey angular spots to the fifth segment*; venter pure white. Legs pale bronzy-brown; femora white beneath; hind femora entirely so.

FEMALE.- *Head* with the flat scales uniformly olivaceous-black with dull bronzy reflections, with a well-defined continuous margin of white ones, broadening towards the base; there is also a napal patch of loose flat dusky-white scales. and below them a few narrow-curved, and very short upright forked ones; clypeus black. *Palpi* shining bronzy-black. *Proboscis* with bronzy-brown and ochreous scales, basal half with a median, interrupted, line of white scales. *Antennae* with the basal segment greyish-black, second segment pale brown, remaining segments darker; hairs dark brown or black, pubescence white. *Thorax:* Prothoracic lobes clothed with pure white scales; mesothoracic scales in front narrow-curved, white; at the sides from the base of the wings numerous flat, loose, white ones; dorsally the whole area is sparsely clothed with narrow-curved dark-brown scales intermixed at the sides and posteriorly with a few greyish ones; scutellar scales dull white, with a few shorter black ones at the base of the mid lobe, and also at the base of the marginal hairs to the lateral lobes. *Abdomen* with the first four segments brownish-black; a few dull white scales on the base of the fourth indicating the presence of a more or less imperfect basal band; venter uniformly

* Remaining segments wanting.

white as far as the end of the fourth segment; the remaining segments wanting. *Legs* with the coxae dusky brown, paler apically, with numerous flat white scales; scales to anterior and mid-femora smoky-brown above, ventrally they are white; hind femora clothed with white scales, except a narrow dorsal and a broad apical anterior patch of blackish scales; tibiae and tarsi of anterior and mid-tarsi blackish; posterior tibiae with a broad apical white band, tarsi wanting. *Wings* with dark bronzy-brown scales, those on the costa much the darkest, almost black; first submarginal cell much longer than the second posterior; posterior cross-vein nearly three times its length from the mid.

Length about 4 mm.

Locality:—Only two specimens of this species were caught, during the day, in a European's house at Kasongo (Dec.).

Scutomyia sugens, Wiedemann

Localities:—Matadi; Leopoldville (Dec. and March); Kisui.

Circumstances of capture:—Adults were caught in the bush at some distance from houses. They were seen to attempt to bite at mid-day.

Breeding places:—One adult was reared from a larva taken from a puddle on clayey ground containing clean rain-water.

Catageiomyia senegalensis, Theobald

A culicid with the habits of this mosquito was observed at Banana. No specimens were taken, but it is believed, from a hasty field examination, that they were referable to this species.

DUTTONIA, nov. gen. (Newstead)

FEMALE.—*Head* clothed principally with flat scales, with narrow-curved ones behind and a *row bordering the upper half of the eyes;* upright forked scales on the dorsal area. *Mesothorax* with small, narrow-curved, and minute hair-like scales; scutellum with flat scales, *the lateral lobes small, somewhat tuberculate, and furnished with eight bristles. Palpi* short. Fork cells long, almost equal; lateral vein scales long and narrow, but broadening towards the apex of the wing.

MALE.—With the anterior tarsi subchelate.

This genus is apparently related to *Aedimorphus* (Theobald), but differs in the presence of narrow-curved border scales to the eyes and the character of the lateral lobes to the scutellum. The number of bristles to the small lateral lobes are also unusual; and moreover there is a complete absence of flat scales* to the mesothorax.

Duttonia tarsalis, n. sp. (Newstead)

(Pl. ii, figs. 6-8)

Head grey. Thorax rich dull orange-brown with two median pale lines. Abdomen dark brown with silvery-white basal bands; apical segment almost entirely white. Legs pale ochreous-yellow, scales on tibiae and tarsi pale brown, femora faintly speckled.

MALE.—Lateral and median flat scales of the *head* dull white; the

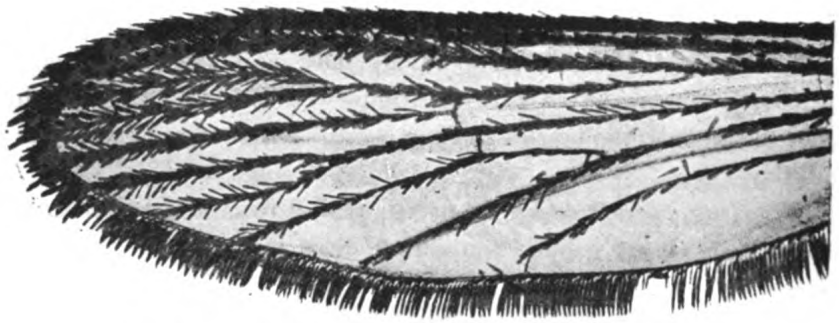


FIG. 2.—*Duttonia tarsalis*. WING OF MALE. × 40.

narrow-curved ones bordering the eyes pale yellow; napal group of narrow-curved scales pale yellow, in some lights almost white; upright forked scales in front yellow, napal ones black, lateral ones yellow. *Palpi* of four segments, apically furnished with long hairs, pale ochreous with dark brown scales and indications of a faint median band. *Proboscis* dark brown. Segments of *antennae* dusky white; nodes dark brown; hairs silvery-grey and pale brown. *Thorax* deep rich orange-brown,† with a few scattered white scales, numerous minute, almost hair-like, jet black scales, and a few golden-yellow ones in front

* Theobald (Mono. Culicid., Vol. III, p. 290), in his description of the genus *Aedimorphus* says there are no flat scales on the thorax. In the type of *Ae. domesticus* the anterior white spots on the mesothorax are undoubtedly formed of small flat scales of the same size and character as those on the scutellum.

† One of four males has the thorax dark brown.

and towards the scutellum; the yellow scales also form two lateral spots in front of the wings; there are traces also of a lateral white line extending from the wings to the front of the mesothorax; scutellum with flat white scales; pleurae dull ochreous with patches of white scales. *Abdomen* very pale ochreous, with dark brown and dull ochreous scales intermixed; basal bands of pearly-white scales; bands 3 to 5 broader than the rest; terminal segment almost covered with pearly-white scales; venter pale ochreous with broad indefinite, basal, dusky-white bands. *Legs* pale ochreous, speckled throughout with ochreous-brown scales; anterior tarsi with the terminal segment subchelate; base considerably dilated, ventrally concave; base with four apparently bilateral spines, apex with a short curved spine, and immediately anterior to it a long plumose hair with a bulbous base; ungues subequal in length, the larger with a long central tooth, the smaller with a short basal tooth; mid-ungues unequal, the longest simple, the smallest with a central tooth; hind ungues approximately equal, simple. *Genital armature* with the basal segment of the claspers furnished with numerous long bristles, two of which, near the inner apical extremity, are narrowly lanceolate; second segment broadly dilated at the apex with an outer, curved, spine-like projection; terminal claw small, slender. *Wings* sparsely clothed with small brown scales; first forked vein narrower and slightly longer than the second posterior.

FEMALE.—*Head* dark brown; median flat scales smoky-brown, lower laterals forming a relatively large white patch; median narrow-curved ones yellowish-white, frontals white, marginal series to eyes pale yellow; upright forked scales numerous, apparently all black. *Antennae* very dark brown, with numerous minute, loose, flat, white scales, giving the segments a mealy appearance. *Palpi* pale brown with darker scales; tips with dusky white hairs. Clypeus very dark brown with minute, scattered, white scales like those on the antennae. Eyes yellowish-brown with two to three black spots. *Thorax* as in the ♀, but with a well-marked series of white scales in front of the mesothorax. *Legs* with patches of white scales on the coxae; underside of hind tibiae clothed with white scales, with a broad apical white band; the legs are also more uniformly coloured and browner than in the ♂, but the upper surface of the tibiae are speckled. *Abdomen* clothed with dark brown scales with paler ones at the articulations;

venter with lateral white spots; median area greyish.

Length 4·50 to 5 mm.

Taken in thick bush at noon, October 4th, 1904.

Locality:—Kisui.

Duttonia africana, n. sp. (Newstead)

Head with the palpi and proboscis black. Thorax dark brown in front, paler behind. Abdomen dark brown, paler at the articulations. Legs smoky-brown with pale knee spots; apex of hind tibiae with a white band.

FEMALE.—*Head* clothed principally with flat scales, median and frontal ones blackish, those at the side with dull bronzy reflections, below these there is a narrow curved band of bright ochreous ones,



FIG 3.—*Duttonia africana*. WING OF FEMALE. × 40.

which in some lights appear dusky yellow; there is a large patch of narrow-curved, dull yellow scales behind, and a single series of golden ones bordering the anterior half of the eyes; upright forked scales numerous and scattered over the whole of the dorsal area, black. *Antennae* dark brown; basal segment brownish-yellow at the apex, darker at the sides. *Palpi* short, black, and extremely hairy. *Proboscis* clothed with black scales. *Thorax* shining, rich, dark brown in front, paler behind; prothoracic lobes with large, white, narrow-curved scales and black bristles; mesonotum with minute, blackish, hair-like scales, and a few larger, narrow-curved, yellowish scales; there are also numerous black bristles at the sides and on the dorsum in the

region of the wings; scutellum trilobed; mid-lobe with small, flat, dark brown scales, and traces also of a few white ones; lateral lobes small, almost tuberculate, furnished with eight long bristles, and a patch of flat dull white scales. *Abdomen* clothed with dark brown and almost black scales; venter with broad, pale, basal bands interrupted by a median ochreous line which broadens towards the apex. *Legs* uniformly brownish-black; femora pale beneath; knees and tibio-tarsal articulations pale; hind tibiae with a broad apical white band. *Wings* with the first submarginal and second posterior cells about equal in length; posterior cross-vein about its own length distant from the mid.

Length about 5 mm.

Locality:—A single ♀ taken in a water-closet at Kasongo, April 7th, 1905.

Culex albitarsis, Theobald

Localities:—Boma; Kasongo (Feb.).

Circumstances of capture:—Adults were taken during the day in the open, and at night in a European's house.

Breeding places:—Larvae taken from swamps produced adults in captivity. The pupa of this mosquito is particularly large.

Culex annulioris, Theobald

All four specimens of this species were obtained from larvae caught in a grassy swamp near Leopoldville.

Culex dissimilis, Theobald

Specimens were caught in a tent near Tumba, October 26th, 1903.

Culex fatigans, Weidemann

Localities:—Boma; Matadi; Wathen; Leopoldville (Dec.); Mswata; Irebu; Coquilhatville; Bamamia; Lisala; Basoko; Stanley Falls; Kisui; Kasongo (Feb.).

Circumstances of capture:—Imagines were taken by day and by night in the dwelling-places and in the mosquito nets of both Europeans and negroes. They were also caught at noon on steamers

and in thick forest at some distance from any village. At night they were often seen on board the Expedition's steamer.

Breeding places:- Larvae and pupae were found breeding in old tins, in water reservoirs, amongst grass at the edges of rivers, in swamps and in the foul-smelling pools used for steeping manioc.

Field notes:- Enormous numbers of this mosquito, together with *Mansonia uniformis* and its var. *africanus*, were found during the day time in the prison at Bomba. They rested thickly clustered and motionless on the whitewashed walls of the cells, a few inches above a drain used as a urinal by the prisoners; scarcely any of these mosquitoes were seen on the neighbouring tarred portions of the wall. No breeding places existed within 200 yards of the prison.

Culex luteolateralis, Theobald

Localities:- Boma; Bamu Island; Kasongo (March); Lusambo.

Circumstances of capture.- Specimens of this beautiful little culex were caught in the early morning and afternoon, in the forest, in coffee plantations and in the houses and mosquito nets of native servants. They attempted to feed but rarely.

Breeding places:- Larvae were taken from amongst the grasses at the edges of rivers and from swamps. We were successful in obtaining imagines from them.

Culex metallicus, Theobald

Localities:- Miambwe; Lusambo.

Circumstances of capture:- Adults were taken in the bush, near marshy ground and at a few hundred yards from the nearest village.

Culex thalassius, Theobald

A single specimen of this species was obtained from a larva taken from a tub on the mission station at Wathen.

Culex tigripes, Grandpré

Localities:—Boma; Wathen; Leopoldville (Dec.); Tshumbiri; Yambinga; Kasongo (April to Feb.); Miambwe; Lusambo.

Circumstances of capture:- Imagines were caught in forests and marshes, during the day time on board the Expedition's steamer and at night in the houses of Europeans.

Breeding places:—Larvae and pupae, allowed to develope in the laboratory, were taken from among the grass at the edges of rivers, from swamps, from the foul water of stagnant puddles, from collections of rain-water in old tins, and from the foul-smelling pools used for steeping manioc.

This is evidently a variable species. None of the females agree with the typical form in the specific coloration of the head and thoracic scales. In the majority of the females, which agree best with Theobald's* description, the narrow-curved head scales are creamy-white in colour with, in some lights, a trace of pale yellow, especially in the frontal and large median ones; and the thoracic scales are smoky-brown with metallic reflections, almost golden in some lights, with the distribution of the yellow spots and lines as in the type. In addition to these variations, five of the females also possess a distinct median band to the proboscis and other distinctive characteristics. These forms are described below as a distinct variety.

Culex tigripes, var. *consimilis*, n. var. (Newstead)

FEMALE.—*Head* with the narrow-curved scales creamy-white, in some lights with a faint trace of yellow; upright forked scales black with smoky-grey tips; median hairs golden yellow. Thoracic scales chiefly of a greyish colour, with dark brown ones intermixed. *Proboscis* brownish-black *with a broad and well-defined median band* of pale ochreous scales; labella pale ochreous-brown. *Palpi* black or brownish-black with a patch of pale bright ochreous scales at the articulation of the first long segment; apex with dusky white scales. *Abdomen* dark brownish-black with scattered pale brown scales and narrow basal bands; basal segment with two median black apical spots; sixth and seventh segments with two lateral apical pale spots. *Legs* as in the typical forms.

Length 6·7 mm.

Localities:—Tshumbiri (July); Kasongo (Feb., April, May); Yambinga (Sept.); Miambwe; Leopoldville. The example from Leopoldville was bred from a larva procured in a grass swamp near the terminus of the railway. It is important to note that three females and four males of the more typical forms were bred from larvae obtained at the same time.

* Mono. Culicid. II, p. 34, 1901.

Culex viridis, Theobald

Localities:—Boma; Wathen; Leopoldville (Dec., 1903); Coquilhatville; Miambwe; Kasongo (Jan.); Lusambo.

Circumstances of capture:—Imagines were caught during the day in the bush, in the mosquito nets covering the beds of "boys," in houses of Europeans and Africans, and in the state rooms of a steamer on the Upper Congo.

Breeding places:—Larvae and pupae were taken from among the grass along the edges of rivers, in swamps, in dirty, foul-smelling collections of water, such as pools used for steeping manioc, and in water collected in old tins. Imagines were reared from these.

Field notes:—This mosquito has been repeatedly observed to have a distinctly green colour during life.

Culex laurenti, n. sp. (Newstead)

Head grey; thorax brown, abdomen paler; legs uniformly pale brown; venter and pleurae, pale, dull orange-brown; proboscis unbanded.

FEMALE. *Head* with the narrow-curved scales pale silvery-yellow in front, the rest white; flat scales white; upright forked ones all black. *Antennae* dark brown, pubescence silvery-grey, in some lights with a faint yellow tinge. *Proboscis* clothed with brown scales, and towards the apex a few scattered, pale ochreous ones. *Palpi* very short; rich brownish-yellow; scales brown. *Thoracic* scales dull golden-yellow; pleurae with a few small groups of narrowly-rounded white scales, some of which are practically spindle-shaped. *Abdomen* uniformly pale brown. *Legs* uniformly brown above with pale brown scales, silvery-grey beneath, in some lights; coxae and trochantae rich brownish-yellow with dark brown scales; no knee spots and no bands to tarsi.

MALE.—*Head* scales as in the ♀. *Palpi* long, pale ochreous-brown with pale brown scales, apical segments dark brown densely clothed with long hairs. *Antennae* with silvery brown hairs. *Thorax* with a few white scales opposite the insertion of the wings; the rest, including the pleural scales, as in the ♀. *Legs* pale ochreous beneath, darker above, with pale brown scales; anterior and mid femora with white scales at the sides, posteriorly. *Abdomen* pale brown with

rather dark brown scales; segments 5, 6, 7 and 8 with a few lateral white scales; terminal segment with a broad apical band of creamy white scales; venter pale ochreous, scales pale brown and white intermixed. *Wing* scales dark brown on the basal half of the veins; apical portion with pale ochreous scales.

Locality:—Leopoldville (Oct. and Dec., 1903).

Two males and one female only were taken.

This species is dedicated to the memory of the Botanist, Professor Laurent, who died while on an expedition to the Congo Free State.

Culex par, n. sp. (Newstead)

(Pl. i, fig. 11)

Head and thorax grey. Abdomen dark brown; fifth and sixth segments with lateral pale spots; venter greyish, dark brown basally. Legs brown; femora and tibia freckled, the former much more so than the latter; anterior and mid tarsi with three narrow pale bands at the articulations. Proboscis with a broad median band.

FEMALE.—*Head* with the narrow-curved scales creamy-white; upright-forked ones pale ochreous, with a few very dark brown ones at the sides; flat scales few in number, creamy white. *Antennae* wanting. *Palpi* short; apical segment minute, clothed with dark brown scales. *Proboscis* dark bronzy-brown with a broad median creamy-white band; labella paler. *Thorax* dark brown; scales chiefly pale ochreous and creamy-white with a few black ones intermixed posteriorly and on the scutellum, the black ones predominate; there are also indications of four small equidistant black spots. *Abdomen* clothed with very dark brown almost black scales; a few isolated dull creamy-white ones on the first and second segment, a small lateral apical spot on the third, a large spot on the fourth and fifth; remaining segments rubbed; venter with a median pale line; last two segments with dull white basal bands, very pronounced on the penultimate segment; the remaining basal segments are not sufficiently clear to determine. *Legs* uniformly pale brown or ochreous-brown; knee spots faintly indicated; femora and tibiae with pale freckles; articulations of front and middle tarsi with three narrow pale bands; hind tibiae somewhat paler than the rest. *Wings* with pale and somewhat metallic-brown scales; fork cells

with their bases almost opposite; posterior cross-vein about one and a fourth times its length from the mid.

Allied to *C. thallasius* (Theobald), but differs in the absence of basal abdominal bands, the broader band to the proboscis, the predominating pale upright forked scales of the head, and also in the colour of the thoracic scales.

Locality:—A single ♀ taken at Tshumbiri on July 5.

Taeniorhynchus fuscopennatus, Theobald

This mosquito was caught only, near marshy ground, in the bush at Lusambo.

Taeniorhynchus annettii, Theobald

Localities: Wathen; Coquilhatville; Kalombe; Lusambo.

Circumstances of capture:—This mosquito was only caught in the bush, and as a rule in the neighbourhood of marshy ground. Its larvae were not seen.

Taeniorhynchus aurites, Theobald

Localities:—Tumba; Tshumbiri; Irebu; Kalombe; Lusambo.

Circumstances of capture:—This mosquito was only caught in the bush, and as a rule in the neighbourhood of marshy ground. Its breeding places were not found.

Taeniorhynchus tenax, Theobald

Localities:—Boma; Leopoldville (Dec.); Lusambo.

Circumstances of capture:—Adults were caught in the bush near marshy ground.

Breeding places:—Larvae and pupae, collected from among the grasses at the edges of rivers and from swamps, produced imagines in the laboratory.

Mansonia uniformis, Theobald

Localities:—Boma; Prince's Island; Leopoldville (Dec., June); Bamu Island; Lulanga.

Circumstances of capture:—Adults were taken in the dwelling-places of Europeans and Africans, in the mosquito nets of "boys," and in the bush.

Breeding places:—Larvae of this mosquito were not seen.

Mansonia uniformis, var. africanus, Theobald

Localities:— Zambie; Boma; Prince's Island; Tumba; Wathen; Leopoldville (Dec. and May); Bamu Island (Jan.); Telegraph Post No. 4; Tshumbiri; Bolobo; Irebu; "in the bush above Lukolela"; Yumbie; Bamamia; Bumba; Basoko; Kama; Benaburungu; Nyangwe; Kasongo; Kalombe; Tshofa; Lusambo.

Circumstances of capture:—This species is by far the commonest mosquito in the Congo. It is seen almost everywhere in the bush, and lives equally well in grass country or forest. It always bites fiercely and feeds with equal readiness by day or by night. At Bamu Island, near Leopoldville, it was actually observed to pierce through canvas-seated chairs and even through the soft goat-skin leather of Madeira boots. It was noted in the diary of the Expedition as very unusual that no species of *Mansonia* were seen in the bush where camp was pitched for the night on the banks of the Congo a short distance above Lulonga.

Adults were caught in the houses of Europeans and Africans in the day time and at night. They were taken from mosquito nets covering the beds of natives and whites and were captured during the day on steamers, in the bush and on sand banks.

Breeding places:—We were not successful in rearing the imago from the numerous larvae we had from time to time in the laboratory.

Melanoconion rimus, Theobald

Localities:— Boma and Kasongo (Dec.).

Circumstances of capture:—The specimens in the collection were all bred from larvae.

Breeding places:—Larvae were taken from among the aquatic plants at the edges of a river and from a disused pit, in a brickyard, filled with water and overgrown with weeds.

SUB-FAMILY AEDEOMYINÆ

Aedeomyia squammipennis, Arribalzaga

Localities:—Boma; Leopoldville (Oct.)*; Yambinga.

Circumstances of capture:—The specimens in the collection were bred from larvae or caught in the evening on board a river steamer.

* No specimen; recorded from field notes.

Breeding places:—Larvae were taken from a grass-grown puddle of clean water.

Uranotaenia balfouri, Theobald

A single ♀ of this beautiful species was bred at Kasongo, December, 1904, from a larva taken in an "old brick pool 15 feet across." Theobald* states that the mid lobe of his type was ribbed, in this specimen it is clothed with scales. It may be important to add that the scales at the base of the wing are of a delicate pale blue; but in certain lights appear white as described in the type.

Mimomyia uniformis, Theobald

This mosquito was observed only at Boma; the specimens in the collections were hatched in the laboratory from pupae collected in a papyrus marsh and from among aquatic plants at the edge of a river.

Field notes:—The pupae are yellow and possess very long and conspicuous respiratory siphons.

Mimomyia africana, n. sp. (Newstead)

(Pl. i, fig. 4)

Uniformly dark bronzy-brown; legs paler.

FEMALE.—*Head* very dark brown, completely covered with flat paler brown scales, no upright-forked ones visible. *Antennae* dusky ochreous, basal segment darker and not scaled. *Palpi* very short, clothed with very dark brown scales. *Proboscis* (imperfect) dark brown. *Thorax* very dark brown, slightly shiny, with numerous long bristles and minute narrow-curved scales, of the same colour as the cuticle; scutellum with a few narrow-curved scales; pleurae with a dark brown central area covered with creamy-white scales; metanotum nude. *Legs* uniformly dark brown, with paler coppery reflections, nodes pale brown. *Wings* with all the veins clothed with brown scales; fringe paler; first submarginal cell narrower than the second posterior cell; posterior cross-vein about its own length distant from the mid cross-vein.

Length about 3·50 mm.

Described from a single female which, unfortunately, has the abdomen and tip of the proboscis imperfect. Apart from this,

* First report Wellcome Research Laboratories, p. 82, pl. vi, fig. 6.

however, the insect possesses well-marked specific characters and can easily be recognised from any other known species by its uniform dark brown colour.

Locality:—Taken at Nouvelle Anvers, on August 14th, 1904.

Mimomyia malfeyti, n. sp. (Newstead)

(Pl. i, figs. 1-3)

Head dark brown, with pale ochreous scales. Thorax dark brown, with bright blue reflections and clothed with long backward-curved bristles; pleurae pale ochreous. Abdomen brown; apical segment paler; venter ochreous. Legs dark brown, with pale ochreous femora. Wings with a pale spot at the base.

FEMALE.—*Head* brown, covered with rather loose, flat, cream-coloured scales and a napal group of black, upright-forked ones. *Antennae* deep brown; basal segment paler. *Proboscis* swollen apically, but much less so than in the ♂, dark brown; labella, dull ochreous. *Thorax* dark brown, shining, *with blue reflections in*

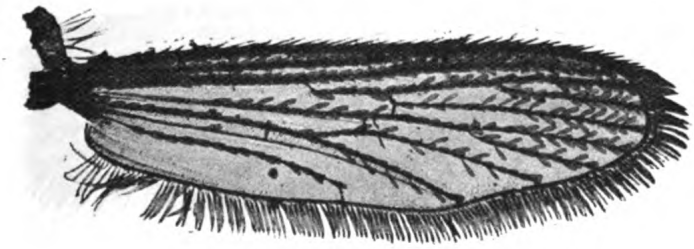


FIG 4.—*Mimomyia malfeyti*. WING OF FEMALE. × 40.

certain lights, but this character is much less evident under the microscope than under a pocket lens; hairs dense and long, especially at the sides above the pleurae; there are also one dorsal and two, more or less distinct, sub-dorsal rows of shorter hairs between which is a double series of *minute*, narrow-curved, ochreous scales, which are very difficult to see *and are often wanting*; scutellum with four or five narrow-curved, almost hair-like, black scales often wanting; metanotum pale brown, *nude*. *Wings* with brown scales and a basal, pale, nude patch; median vein scales *in a double row* on the subcostal and first longitudinal vein for two-thirds of the basal

portion; in single rows on the remaining veins; outstanding scales present on all but the sixth vein, but are most numerous on the apical half of the wing; first submarginal and second posterior cells about equal in length, the former slightly the narrower; posterior cross vein a little more than its own length distant from the mid cross vein. Halteres creamy; knobs clothed with flat brown scales. *Abdomen* unbanded, pale brown or ochreous, with somewhat scattered, flat, brown scales which give it a more or less mottled appearance; apical segment paler; venter ochreous, with four or five dark marginal triangular spots. *Legs* uniformly brown with pale reflections, except the ventral and basal half of the femora, which are pale ochreous; all the articulations are pale.

Length 2·50 mm.

MALE.—*Head* with dusky ochreous scales. *Thorax* as in the ♀; metanotum darker. *Antennae* densely plumose, dusky brown; basal segment paler. *Proboscis* much swollen from beyond the middle

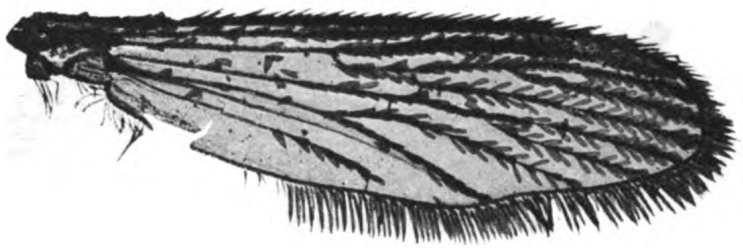


FIG 5.—*Mimomyia malfeyti*. WING OF MALE. × 40.

apically; clothed with dusky ochreous scales; labella slightly darker. *Palpi* long, thin, apparently of three equal segments, tip reaching just beyond the base of the swollen portion of the proboscis. *Abdomen* as in the ♀ but if anything more distinctly mottled. Anterior tibiae with a long, outer, simple spine, and an inner, tridentate falcate spine. *Genital armature* with the basal segment stout; arising from its inner surface, at the base, are four long spinose hairs; second segment about equal in length to the first; terminal claw short; herpes relatively short, with one long and two minute spines.

Length 2 to 2·5 mm.

This species comes very near to *Mimomyia uniformis* (Theobald); but differs in the colour of the head scales, in the presence of a double row of vein scales to the costal and first longitudinal vein, and in the thorax with its regular series of bristles and bright blue reflections, and also the mottled appearance of the abdomen.

The peculiar proboscis and palpi of the ♂ are characteristic also of *Mimomyia uniformis*.

Locality:—This mosquito was observed only at Boma; the specimens in the collection were hatched in the laboratory from larvae taken in a grass-grown puddle of clean water.

This species is dedicated to Major Malfeyt, Haut Commissaire Royal, in recognition of constant courtesy and assistance rendered during the expedition.

POSITION UNCERTAIN

Neomelaniconion palpale*, n. sp. (Newstead)

(Pl. i, figs. 7-9)

Uniformly brown. Antennae densely plumose; hairs smoky-brown with the apical portions grey; segments clothed with minute white scales; nodes black.

MALE.—*Palpi* long, dark brown, with a few scattered blackish-brown scales; apex faintly clavate and densely clothed with long brown hairs, more especially so at the sides, where they form a long continuous fringe. *Proboscis* straight, pale brown, clothed with dark brown scales; apex extending to base of apical segment of antennae. Central area of *head* clothed with narrow-curved, pale golden-yellow scales, intermixed at the sides with long, flat, dusky-brown scales; sides with flat dark brown scales intermixed with a few yellow and dull cream-coloured ones; nape with a few small, upright-forked, black scales. *Mesothorax* (partly denuded) and scutellum with narrow-curved pale golden-yellow scales; metanotum nude; prothoracic lobes denuded; pleurae with small dull cream-coloured scales. Halteres pale ochreous basally, knobs pale brown, with a few small, flat, brownish scales. *Abdomen*, where denuded, almost black; scales pale brown with faint, dull, greenish-blue and dull coppery reflections; two well-defined sub-median, triangular, black spots at the base of the third

* Theobald, M.S.

segment, and there are traces of similar markings on the second and fourth; fifth and sixth, with a basal band of pale yellow; venter, at the base, with pale golden-yellow scales in the centre, dull creamy ones intermixed at the sides; the remaining segments are rendered invisible by the curved condition of the abdomen. *Wing* scales uniformly brown, darker on the costa; posterior cross-vein about one and a half times its length distant from the mid cross-vein; first submarginal cell scarcely longer than the second posterior, the latter with the veins widely divergent; outstanding scales long, narrow, sides parallel, apex convex; fringe scales long, lanceolate. *Legs* uniformly brown; posterior tibiae with a pale apical band; tarsi dark brown.

Length about 2·50 to 3 m. The specimen has the abdomen curved under, so that it is impossible to give the exact measurement.

Locality:—One male bred from a pupa taken in a pool on an island below Basoko, September 3, 1904.

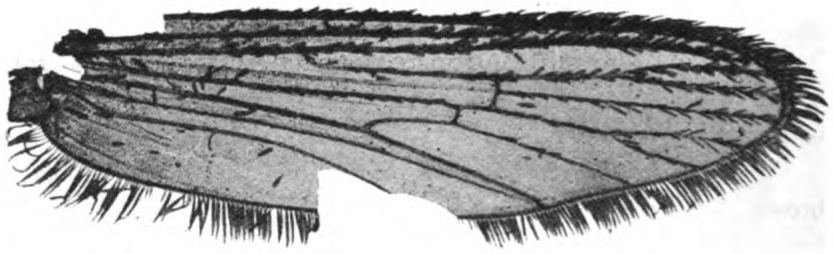


FIG. 6.—*Neomelaniconion palpale.* WING OF FEMALE. × 40.

Anisocheleomyia quadrimaculata, n. sp. (Newstead)

(Pl. i, figs. 5, 6)

Head pale brown with narrow ochreous margins. Antennae dusky. Proboscis swollen at tip; black with scattered bright ochreous scales; labella bright ochreous. Thorax bright ochreous with four black spots, the anterior pair being much the largest; behind the second and smaller pair is a large and somewhat ill-defined black band. Abdomen black with a dorsal series of elongated pale spots. Legs spotted and banded; femora dilated apically. Wings with scattered black scales, those along the costa forming a basal bar and two spots.

FEMALE.—*Head* with both black and pale cream-coloured upright-forked scales, the latter predominating; anterior portion with rather large, narrow-curved, bright fulvous scales, and apparently a few long flat ones, intermixed with the former are a few black ones tipped with bright fulvous; sides with flat, somewhat loose, cream-coloured scales. *Antennae* dusky brown; nodes pale. *Palpi* densely scaled, black, dorsally with white tips. *Proboscis* swollen apically; scales black intermixed with a few cream-coloured ones; swollen portion entirely black; labella bright ochreous. *Legs* with the anterior femora pale ochreous with a black apical patch in the centre of which is a distinct central crescentic band; mid and hind femora black, the former with two, the latter with one yellowish band; tibiae black, each with a sub-apical but somewhat diffused band; tarsi black with bronzy-brown reflections, each with five distinct yellowish bands; ungues equal, simple. *Wings* with bronzy-black and cream-coloured scales; basal half of costa with a long black bar and two irregular spots; all the nervures with irregular groups and isolated black scales; fringe pale; costal scales bronzy-black, becoming paler towards the apex; out-standing scales claviform; a few of the black ones, especially those on the sixth vein, heart-shaped. Halteres cream-coloured with dark brown scales at the tips. *Thorax* with narrow-curved, golden-yellow scales; four lateral spots of black ones, the anterior pair are the largest, and there is a broad indefinite band behind the second pair; scutellum with pale ochreous narrow-curved scales; metanotum black, with three dusky-yellow lines, and a basal, linear, patch of minute, flat, cream-coloured or white scales; prothoracic lobes and pleurae with flat, dull, cream-coloured scales. *Abdomen* clothed with black scales, having brownish coppery reflections; each segment with a long narrow median, basal, patch of dull cream-coloured scales; apical segment only with two lateral pale spots; venter ochreous.

Length 3·75 mm.

Locality:—Boma; bred from a larva which was caught in a marshy pool.

BOYCIA, nov. gen. (Newstead)

Head with a median area of narrow-curved scales, flat, loose, lateral ones, and numerous upright-forked ones. Thorax and

scutellum with narrow-curved scales. Wings with the fork cells relatively short; anterior forked vein slightly shorter than the first posterior; scales resembling those in *Mimomyia*. Palpi short in the ♀, long and clavate in the ♂. Proboscis swollen at the tip in both sexes. The narrow curved scales in the mid region of the head are fewer in the ♀ than in the ♂; and in the former the central ones are arranged in two distinct lines, but are almost completely hidden by the numerous upright-forked scales.

This genus is somewhat difficult to place, but agrees best with the group of Culcinae in which the palpi of the males are swollen at the tips.

Boycia mimomyiaformis, n. sp. (Newstead)

(Pl. ii, figs. 1-3)

Head greyish. Mesothorax grey-brown, with two anterior spots and a broad transverse band opposite the insertion of the wings, black. Abdomen black, with pale narrow basal bands; venter ochreous; pleurae pale ochreous. Legs brown; base of femora and tarsi (in certain lights) paler. Proboscis swollen towards the apex.

FEMALE.– Central area of ***head*** with narrow-curved, creamy-white scales, those in the centre forming two longitudinal series, intermixed with these are numerous creamy, upright forked ones, extending almost to the front of the head; sides with large, loose, spatulate, creamy scales; nape with numerous upright-forked, black scales; all the forked scales are broadly dilated with, usually, five well-marked dentitions. *Thorax* densely clothed with narrow-curved pale golden-yellow scales; two anterior spots and a broad transverse band of black ones; the band faintly constricted in front giving it, in some specimens, the appearance of two large confluent spots; scutellum with large, narrow-curved, creamy-white scales, those on the lateral lobes with golden reflections; metanotum nude*; pleurae creamy-yellow. *Abdomen* clothed with dark brown and black scales, appearing bronzy-purple in some lights, and narrow basal bands of pale ochreous scales; venter pale ochreous, with lateral, angular, dark brown, spots. *Legs* unbanded, bronzy-brown; femora pale ochreous-

* Two females show a single curved scale, but these are apparently loose ones from the metathorax.

brown at the apices, forming pale knee spots; tibio-tarsal articulations pale; last three segments of tarsi bronzy-ochreous.

Length 2·50 mm.

MALE.—*Head* with a well-defined median, long, angular patch of narrow-curved creamy scales, no regular series as in the female; sides broadly clothed with large, rather loose, flat, cream-coloured scales, and a rather large lateral patch of brown ones; a few short upright forked ones at the nape, black, and a few, very short, creamy ones towards the front of the head. *Antennae* densely plumose, hairs grey with a few black ones intermixed. *Palpi* strongly clavate, with a pale cream-coloured band at the anterior end of the first long segment; the remaining portion of the segment and the basal half of the succeeding one with very dark brown, almost black scales; apical

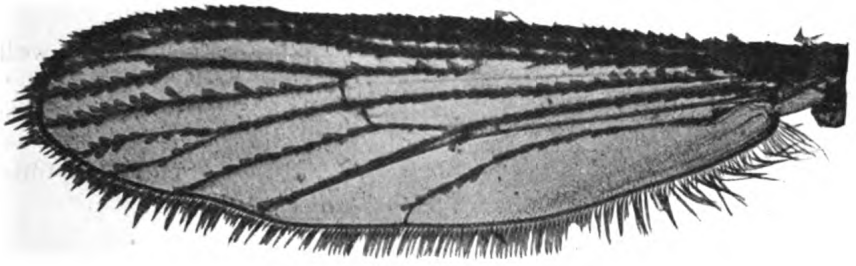


FIG. 7.—*Boycia mimomyiaformis*. WING OF FEMALE. × 40.

segment with short stiff yellow bristles. *Proboscis* swollen apically, dark brown; labella paler, especially at the tips. *Mesothorax* with the narrow-curved scales dusky yellow intermixed with grey and a few black ones, and there are two ill-defined lateral black spots in front of the wings; space between the wings denuded, dark brown; pleurae and legs as in the ♀. *Wings* with pale brown scales; anterior forked vein slightly shorter than the first posterior, the latter much the widest; both are relatively short. *Abdomen* as in the ♀ but the bands are a little more pronounced.

Localities:—All our specimens of this mosquito were bred from larvae caught at Boma (Oct.) in a papyrus swamp and among the aquatic plants of a small stream.

FAMILY CHIRONOMIDÆ

Ceratopogon sp. ?

This fly was only twice observed, once at Binza (Dec.), near Leopoldville, and again near Tumba. In these localities it was the most common bloodsucker observed. Although it is almost invisible its bite raises a large weal which itches for hours.

Chironomus sp. ?

The specimens which we obtained of this insect were bred from larvae, at first sight resembling mosquito larvae, caught in a swamp at Boma. This insect was also seen at Leopoldville.

FAMILY PSYCHODIDÆ

Phlebotomus n. sp. ?

The only specimens which we now possess are not sufficiently well preserved for descriptive purposes.

Localities: Leopoldville (March); Wanie Numbu.

Circumstances of capture:—Specimens were taken in the daytime in the bush and in rest houses for Europeans.

FAMILY SIMULIDÆ

Simulium damnosum, Theobald

Localities: Matadi; Lutete; Leopoldville (Nov.); near Miambwe.

Circumstances of capture: Specimens of this species were caught both near, and at some distance from, water. They bite freely.

Simulium sp. ?

Localities: Leopoldville (Nov., 1903, to Feb., 1904); M'Swata; Batikalela.

Circumstances of capture: These flies were caught both near, and far from, water. They sometimes occur in swarms and are often found in houses. They bite freely, are persistent in their attacks, and when crushed emit a peculiar "bed-bug like" odour.

HABITS AND STRUCTURAL CHARACTERS OF THE LARVA OF SIMULIUM*

As the characteristics of the larvae of the European species of *Simulium* are not likely to differ very materially from their tropical relations the subjoined notes may prove of some guidance to students in other parts of the world. The notes refer to a single species, *Simulium ornatum*, Mg.

Shallow *rapid* streams *fully exposed to the sun* on hill sides, moors and open spaces in woods are the favourite resorts of these curious larvae. They are invariably found where the stream flows

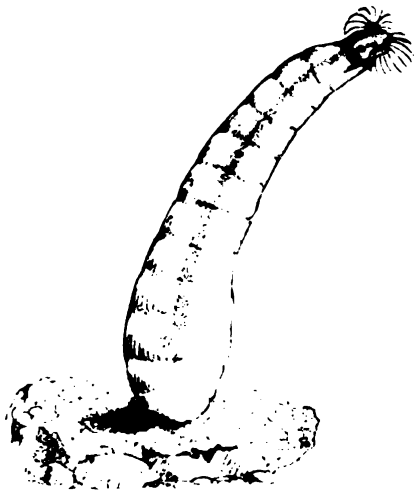


FIG 8.—LARVA OF *Simulium ornatum* in semi-erect position with the fan-like or fringed appendages expanded. × 6½.

most rapidly; never in the side pools or in comparatively still water; and very rarely in those portions of a stream which are overshadowed by trees or shrubs. In streams where all the necessary conditions are available the larvae sometimes occur in countless numbers, covering the *undersides* of nearly every submerged leaf or blade of grass, and also, where not too much overshadowed by aquatic plants, on the stones and fragments of rock at the bottom of the stream.

* These notes were compiled from observations made in England and Belgium in the summer of 1906.—R.N.

When massed together on the stones and rocks they look at a short distance remarkably like a waving mass of dark brown or blackish algae or moss; but this resemblance is much less marked in colonies attached to the leaves and stems of plants.

The larva, when at rest, attaches itself to a fixed object by means of its anal sucker, standing in a semi-erect position (fig. 8) with its head pointing down stream; but in very rapid rivulets the body lies almost prone with the object upon which it rests, or the extremities may be brought together so that the body forms a distinct

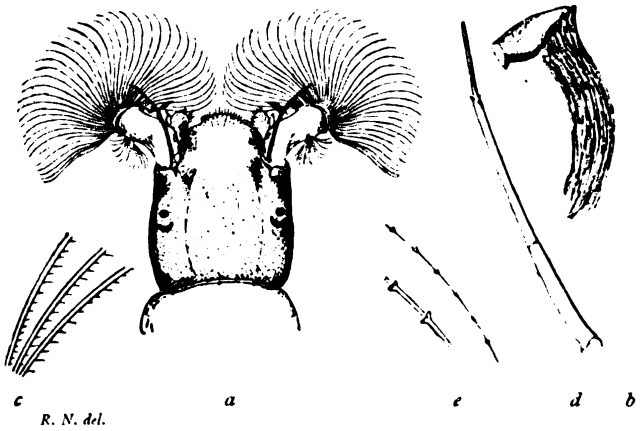


FIG 9.—LARVA OF *Simulium ornatum*:—*a*. head of larva with the fringed appendages expanded, × 40; *b*. fringed appendages closed; *c*. portions of the fringe showing lateral cilia, × 600, about; *d*. antenna, × 150, about; *e*. silken threads showing the characteristic nodes (enlarged).

loop. The head is provided with a pair of beautiful fan-like processes with which the animal sweeps the particles of food into its mouth. Unlike the larvae of mosquitoes they have little or no power of swimming, and when migrating from place to place, their method of progression strongly resembles that of the larvae or looper caterpillars of the geometrid moths. As they progress from place to place silken threads are spun in all directions, forming an irregular web or network along which they can travel with great ease. When forcibly dislodged from their resting places they immediately float down stream to distances varying from two to fourteen inches, suspending themselves by a slender silken thread, freshly spun, or by the network of threads

which may have already existed. Many of them will immediately commence to haul themselves back again by walking along the silken thread or threads, and this they accomplish by forming the body into a succession of loops, in the same way as they progress along stones and plants—the mouth or sucker feet grip the thread firmly while the posterior sucker is brought forward until it almost touches the anterior segments—the action being repeated until a place of safety is reached. Many, however, float down stream supporting themselves by their threads until some fixed object is reached. If a leaf or stem supporting a colony of larvae be pulled partly out of the water the larvae will all crawl back again to the stream and usually fix themselves to the submerged portions of the same leaf or stem.

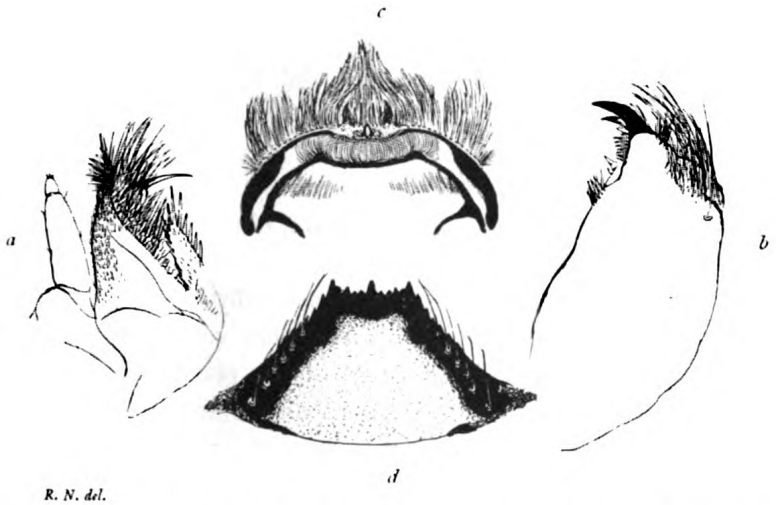


FIG 10.—LARVA OF *Simulium ornatum*:—*a*. maxilla and palpus; *b*. mandible; *c*. labrum; *d*. labial plate. × 125.

If placed in still water the larvae soon become sluggish and death takes place in about 17 hours, but if completely removed from the stream and kept in a thoroughly moist vessel they will survive for a much longer period.

When about to pupate the larva spins for itself a little elongated cocoon, shaped somewhat like the toe of a slipper, with a large opening at the broad end, and this is almost invariably placed with the opening pointing down stream. From this the head of the pupa, with its external respiratory filaments, projects and a sufficient supply of oxygen is thus obtained.

The period of pupation is only of a few days' duration: the minimum being two, the maximum six days. On bright sunny days they begin to hatch as early as 9 a.m. and continue to emerge during the heat of the day. A few examples also hatched in the laboratory during the night, but this was apparently an exception to the rule.

In order to rear the imagines the cocoons should be removed from the stream immediately they are formed; and the leaves and stones to which they are attached should be placed in glass jars covered with fine muslin or chiffon. All that is necessary is to keep the material perfectly moist; however, many imagines were successfully reared from pupae which had become perfectly dry. On no account should the pupae be left in standing water.

On the emergence of the imago the skin of the pupa splits along the median line of the thorax and the fly rapidly pushes itself forwards, being at the same time surrounded by air which gives it a greyish or silvery appearance, especially at the bases of the wings. The moment the insect has freed itself of the pupa case it is instantly carried to the surface when it grips the first object with which it comes in contact in its rapid course down stream, and having gained a foothold instantly takes to wing.* Occasionally the insects were seen completely immersed in the water, where they seemed at perfect ease either when walking along the stems of the plants or cleaning their legs and antennae. In such cases the wings were folded partly round the abdomen so that they tapered to a point behind, and in this way were seen to form a large air cavity. This remarkable trait was observed in still water only.

Description of the larva.—Body cylindrical, swollen posteriorly. Legs in two, coalescent, pairs; the anterior pair are attached to the first thoracic segment and project forward beneath the head and are provided with little hooklets; the second pair (fig. 11) are placed at the anal extremity and form a broad flat sucker which is surrounded by a band of hooklets arranged in regular transverse series.

Head (fig. 9, *a*) relatively large, elongated; labium broadly rounded in front. Eyes represented by two bilateral spots. Antennae (fig. 9, *d*)

* Some species evidently dry their wings before taking flight.
Austen, 'Brit. Blood-sucking Flies,' p. 29 (1906).
Miall, 'Aquatic Insects,' p. 186.

slender, almost filiform, of apparently three segments, of which the first is extremely short, the second about half the length of the third. Immediately below the antennae is a large stout process furnished at the end with a long fan-like fringe of hairs, with a much smaller, but similar group of hairs arising from the centre of the lower lateral margins of the same process. The hairs forming the large fans are provided with an inner lateral fringe of delicate short hairs. *Mandibles* furnished at the tip with two stout and slightly curved spines and long silken hairs. *Maxillae* each with a long slender sub-terminal spine which is sometimes completely hidden in the dense tuft

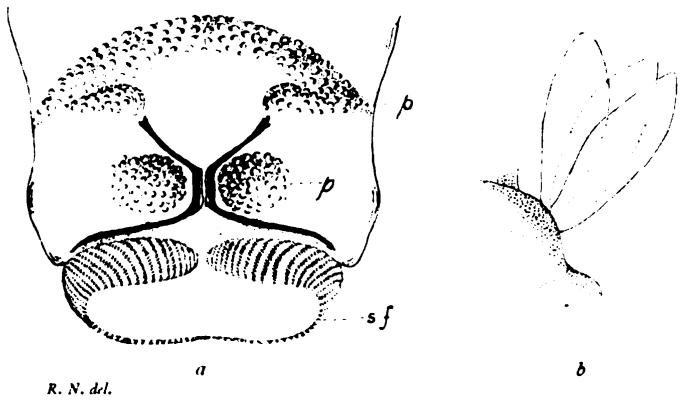


FIG 11.—LARVA of *Simulium ornatum*:—*a*. anal segment; *p*. papillae, *sf*. sucker foot with its coronet of hooks (× 45); *b*. anal appendages (× 45).

of silken hairs. *Antennae* short, stout, of two segments, the terminal one being about one-fifth the length of the first. Labial plate broad and angular; the anterior margin emarginate and furnished with nine strong teeth or spines, the outer teeth bidentate, the median tooth slightly the longest; anterior half of the lateral margins strongly serrate; and there is a submarginal row of long stiff bristles.

Length of fully matured larva 7·50 to 8·50 mm.

FAMILY TABANIDÆ

Hæmatopota duttoni, n. sp. (Newstead)

(Pl. iv, fig. 3)

Face dark brown, clothed with grey pubescence, with two black spots. *Palpi* dull yellowish-brown, sparsely clothed with black

pubescence above, grey at the sides. *Antennae* pale brown, tips black; first segment almost cylindrical, the third slightly longer than the first and second together. *Frontal callus* pale yellowish-brown, attaining the eyes, slightly produced in front; a broad rectangular spot of light rich chestnut between the eyes and a paler but ill-defined spot above it in the centre of the callus; paired spots large, black, and slightly reniform; unpaired spot distinct. No markings on the vertex. *Thorax* dark brown with three equidistant cinereous stripes. *Abdomen* brown, with golden pubescence; first and second segments with several rather large black spots; the remaining segments are spotted only at the sides with the exception of the two last ones, which also bear traces of small spots on the dorsal areas; scutellum dull cinereous. *Legs* pale brown; anterior tarsi and apical half of tibiae almost purple brown. *Wings* with a chequered appearance, apical third darker than the rest; upper rosette fairly distinct.

This species is closely related to *H. similis* (Ricardo), but may be distinguished from it by the presence of a broad rectangular spot between the eyes, the pale brown colour of the frontal callus, the large paired spots not reaching the eyes, and the presence of clypeal spots.

Length 10·50 mm.; wing 10 mm.

Localities: Nyangwe; Kasongo; Tshofa; Miambwe.

Circumstances of capture: This fly was caught along rivers and about cattle.

Hæmatopota trimaculata, n. sp. (Newstead)

(Pl. iv, fig. 2)

Head and thorax brownish-black; abdomen smoky-brown; legs black, tibiae dusky-white or pale ochreous; wings smoky-brown with three, equidistant, oblique dusky-white elongated spots.

FEMALE.—*Head:* Vertex with a deep triangular depression behind; uniformly black, shining. *Proboscis* and *palpi* brownish-black; pubescence greyish-black. *Thorax* finely punctate; dark brownish-black with two sub-median, obscure, narrow brownish streaks; transverse suture at the sides, and posterior angles paler than the rest; pubescence scanty, dusky-yellow. *Abdomen* smoky-brown, the last three segments darker; pubescence scanty, darker brown than that of the thorax; venter scarcely lighter than dorsum. *Legs* with the coxae and femora brown; anterior and hind tibiae ochreous-

white, apices dark brown; mid-tibiae entirely ochreous-white; tarsi black; first segment of posterior tarsi ochreous, the remaining segments brown-black; first segment of mid-tarsi pale at the basal half, the remainder brown, the other segments are wanting. *Wings* narrow, uniformly smoky-brown; with three costal dusky-white spots; the first, about midway between the base and apex of the wing, is placed below the costa and immediately above the first transverse vein; the second about midway between the first and third spots, forms a continuous oblique band extending from the costa to the base of the first forked vein; the third is slightly narrower and extends from the costa to the hind margin, leaving the extreme tip of smoky-brown.

Length 9 mm.; length of wing 8 mm.; greatest width of wing 2·75 mm.

A clearly distinct species, easily recognised by the curious coloration of the wings.

Locality:—Two specimens were caught in a European's house at Yakusu.

Hæmatopota brunnipennis, Ric.

Localities:—Coquilhatville; Nyangwe; Kasongo.

Circumstances of capture:—These flies were only caught on the river.

Hæmatopota spp.

Several rather worn specimens belonging to this genus were taken on cattle and horses at Matadi. Just to the north of the Limposo river one flew into the carriages of a moving train.

Chrysops dimidiatus, v. d. Wulp.

(Pl. iv, fig. 1)

Localities:—Banana (Dr. Etienne); Matadi; N'Kussu; Wathen; Tshumbiri; Lisala (reported by Rev. K. Smith); Yakusu.

Circumstances of capture:—Specimens were taken both near and at short distances from water. One was also caught in a European's house.

Atylotus nigromaculatus, Ric.

This fly was caught in a European house placed on high ground at the mouth of the Gambia River.

Tabanus canus, Karsch.

(Pl. iv, fig. 9)

Localities:— Lutete; Matadi; Lukolela; Ikelemba and Lopori rivers (March and Nov., Major Malfeyt).

Circumstances of capture:— This fly was only taken by persons travelling in canoes. It is reported to fly very rapidly and its bite is said to be very severe.

Tabanus dorsivitta, Walk.

(Pl. iv, fig. 4)

The only specimen we obtained was collected by the Rev. and Mrs. Billington at Tshumbiri.

Tabanus fasciatus, Fabr.

(Pl. iv, fig. 14)

This species is given to somewhat marked variation.

In several of the examples there is an entire absence of the apple-green colour on the abdomen and it is also of a more decided buff-yellow than is seen in the type. Other examples may be considered as transitional between *T. fasciatus* and the sub-species *niloticus*, Austen.

Localities: Matadi; Leopoldville (Nov., Dec.); Lopori River (March, Major Malfeyt); Nouvelle Anvers; Tubila; Kasongo; Basongo; and at many places along the Gambia.

Circumstances of capture: This insect was seen only near water. Most of the specimens were taken while travelling by steamer or in canoes. Two specimens were, however, caught in the houses of Europeans.

Field notes:—While living this fly is a very beautiful object. The eyes are a bright, metallic, pea-green, and the colours of the body are very much brighter than in dried specimens. It flies strongly, and is often seen on board steamers over 100 yards from the river's bank.

Tabanus gabonensis, Macq.

(Pl. iv, fig. 15)

Localities:—Lukolela (July, Rev. Whitehead); Ikelemba and Lopori rivers (March, Nov., Major Malfeyt); Baringa (July, Dr. Angela); Nouvelle Anvers (April, Dr. Müller).

Circumstances of capture:—Most of the specimens were taken by persons travelling on the rivers by steamer or in canoes.

Tabanus gratus, Loew

This species was caught in March, 1903, on the Kunchau Creek, about 175 miles up the Gambia River.

Tabanus par, Walk.

(Pl. iv, fig. 6)

A single specimen of this insect was caught on board a steamer plying on the Gambia River.

Tabanus pluto, Walk.

(Pl. iv, fig. 7)

This Tabanid was caught in the Congo near the river at Matadi and Kisantu.

Tabanus rufipes, Macq.

(Pl. iv, fig. 8)

Localities:—Matadi (?); Tshumbiri (April, Nov., Rev. Billington); Bolengi; Lulanga; Lisala; Stanley Falls; Wanie Numbu; Kasuku; Tshofa; Pania Mutombo; Lusambo.

Circumstances of capture:—This fly was caught only near water, usually while attempting to bite passengers in canoes or steamers.

Field notes:—They fly very rapidly and strongly, and have been seen at over 100 yards from the banks of the river. They seem to be very local. For example, near Pania Mutombo ten or a dozen were seen at the same moment darting about a canoe. A little further down the river towards Lusambo none were seen for miles.

Tabanus tarsalis, Adams

(Pl. iv, fig. 13)

This insect was only seen once, at Lutete.

Tabanus unimaculatus, Macq.

(Pl. iv, fig. 5)

One specimen of this fly was collected in November, 1903, at Matadi by Dr. Bourguignon.

Tabanus alboventralis, n. sp. (Newstead)

Thorax dark brown with three narrow lines; abdomen darker brown, with a bilateral series of oblique white spots. The whole of the ventral surface, including the head, white. Legs pale ochreous, with the tarsi and apices of the tibiae dark brown.

FEMALE.—*Head:* Space between the eyes ochreous-grey, with two broad transverse brown bands, the whole of the under-surface of the head, as well as the posterior surface, clothed with dense white pubescence. *Antennae* pale brown. *Palpi* pure white and wax-like. *Thorax* dull grey-brown, with a median and two sub-median greyish lines; margins grey, as well as the posterior margin of the scutellum. *Abdomen* dark brown, with narrow pale apical bands; the first to the fifth segment with a sub-median, oblique, elongated spot of greyish-white, the spots gradually diminish towards the posterior extremity, so that the last is scarcely visible, while that on the second segment extends right across the segment, terminating at the apical band; there is also a faint median line formed by patches of golden-yellow hairs; venter pale brownish clothed with white powder and pubescence, apical margins to all the segments with a distinct, narrow, ochreous border. *Legs* with the coxae, trochantae and femora white; tibiae pale ochreous with the apical third brown; tarsi all dark brown. *Wings* transparent, without markings.

Length 10 mm.; length of wing 8 mm.

Locality:—Caught during September and October, 1902, in the neighbourhood of Oyster Creek, near the mouth of the Gambia River.

It is closely allied to *T. obliquemaculatus* (Macq.), but Mr. A. E. Austen, to whom specimens were submitted, thinks that it is an undescribed species.

Tabanus billingtoni, n. sp. (Newstead)

(Pl. iii, fig. 1; Pl. iv, figs. 10-12)

Female black; margin of thorax and basal segment clothed with ochreous pubescence; second, third and fourth segments with a narrow pale apical band. Wings with a narrow median dark zigzag

line and a broad sub-apical band; apex with a clear triangular space; the anterior basal, and succeeding cell clearer than the rest. Male: Thorax red-brown, covered with grey dust. Abdomen black, with narrow grey bands to all the segments. Nervures of wing all margined with brown. Fore tibiae in both sexes white; the remaining segments blackish.

MALE.—*Head:* Eyes dull bronzy-brown (in dry specimen); ocelli ochreous; space between the eyes black, shining, brown below; clypeus and cheeks grey, the latter with long, silken, white pubescence; posterior surface of head dull grey. *Antennae* black, basal segment clothed with grey dust, the rest with ochreous-brown pubescence. Proboscis and labium black. *Thorax* in front of transverse suture, dull castaneous, with a narrow, faintly-indicated median line, and two narrower sub-median lines, which terminate in front as two black depressions; margins dusky greyish-brown; suture opposite the sub-median lines orange-brown, continued downwards as a short narrow streak; posterior half of thorax slightly darker than the anterior; the whole is covered with a greyish powder, and scanty black pubescence; scutellum and pleurae grey, but the latter are paler than the former. *Abdomen* smoky-black with narrow grey apical bands to the segments, these gradually diminish towards the apex, until they entirely disappear on the last one; venter of the same colour and banded as on the dorsum. *Legs* black; mid femora dark piceous; upper two-thirds of anterior tibiae dull white, the remaining third black. *Wings* with the nervures dark brown, all with a broad diffuse band of orange-brown surrounding them.

Length 16 mm.; length of wing 15 mm.

FEMALE.—*Head* as in the male, but the face is pale ochreous. *Thorax* rich dark brown, with two, narrow, sub-median pale fulvus lines which gradually darken and entirely disappear at the transverse suture; margins with a well-defined band of pale fulvus pubescence which is continued round the margin of the scutellum, and in front extends over the pleurae. *Abdomen* black, piceous in some lights, basal segment pale fulvus with the fringe of the pubescence paler; second, third and fourth segments with a conspicuous narrow grey apical band; venter with similar bands on the second to the fifth segments. *Legs* as in the male, but the anterior tibiae have the basal half only of a pale ochreous or dull white colour.

Length 17 to 20 mm.; length of wing 16 mm.; expanse of wing 34 to 35 mm.

Localities:—Several females of this species were first sent to us by Rev. and Mrs. Billington, from Tshumbiri. It has received its specific name as a mark of our appreciation of the interest shown in the work of the Expedition by the collectors. A female was also caught, near the river, at Bolengi. The only male we possess was sent to us by the Rev. M. Ave, from near Matadi.

It is an extremely well-marked insect, and is easily recognised from the other African species of *Tabanus* both by the banding of the abdomen and the markings of the wings.

Tabanus spp. incert.

Six additional species of this genus were taken in the localities named below; but the specimens have all suffered from mould and other injuries, so that it is impossible either to identify them with any certainty or to give adequate descriptions of them. One can only add that two of these specimens are referable to the *Socialis* group.

Localities:—Luano on the Kwilu River (Major Malfeyt, June, 1905); Matadi; Wathen, Leopoldville (Dec.); Lukolela (Rev. Whitehead, Aug.); Kuzu, in the region of the Lower Congo.

Circumstances of capture:—Two of the specimens were caught in a European's house.

FAMILY SARCOPHAGIDÆ

Sarcophaga spp.

Localities:—The representatives of this genus were collected at Leopoldville (Dec.); at Lokolengi on the Lopori river (Sept., Major Malfeyt); and at Kasongo (April); but the genus is a very widely distributed one throughout the Congo Free State.

Circumstances of capture:—Specimens were caught in the open and in privies. Adults were hatched in the laboratory from larvae brought by natives in mistake for larvae of *Auchmeromyia luteola*. Some species are viviparous, and their larvae are very rapidly deposited. Within a very few minutes freshly excreted faeces are often almost covered with them.

FAMILY MUSCIDÆ

Pycnosoma marginale, Wied.

Specimens of this beautiful species were collected only at Leopoldville.

Pycnosoma elara, Walk.

This fly was only observed at Leopoldville. The specimens were hatched from larvae brought by natives in mistake for the maggots of *Auchmeromyia luteola*.

Pycnosoma putorium, Wied.

Specimens of this insect were collected at Wathen. It was not seen in any other locality.

Pycnosoma sp. ?

The only specimen we possess is not in a sufficiently perfect condition for verification. It was captured, in company with others, at Tshumbiri.

Lucilia fuscina, Walk. ?

Localities:—Tshumbiri; Kutu; Basoko; Nya Lukemba (Jan., Dr. De Maria).

Circumstances of capture:—These flies were caught in the bush and were hatched from larvae found in the mud floor of a native hut. They are said to follow cattle.

Lucilia spp. ?

Several additional specimens of this genus were caught at Wathen and at Kutu (Feb.).

Auchmeromyia luteola, Fabr.

(Figs. 12-14)

Localities:—San Salvador; Noki; (Portuguese Congo, reported); Lukungu; Matadi; Wathen; Kimfuti; Leopoldville (Oct.-June); M'Swata; Tshumbiri; Lukolela; Irebu; Bikoro (Rev. Clark); Bamamia; Lisla; Upoto; Bongandanga (Rev. Gamman); Basoko; Loeka (reported); Yalembe; Yandongi; Romee; Yakusu; Stanley Falls; Wanie Numbu; Ponthierville; Kirundu; Utikakadjia; Kumba;

Lokandu; Sĕndwe; Nyangwe; Kasongo (Nov. to April); Molemba; Tshofa; Miambwe; Kabinda; Lusambo; Portuguese Angola (Bastian, interpreter at Lusambo); Lake Tchad (Monsieur Chevalier); ? Lagos (native reports); Sierra Leone (reported by Captain Grattan).

Circumstances of capture:—Most of the specimens were taken during the day time in or near the houses of natives and Europeans. One or two were caught at night as they buzzed loudly about a European's house.

Bionomics:—We have little to add to the description of the habits of this fly published in Memoir XIII of this School. The flies were again noticed to be attracted by the sleeping mats of natives. As before, the pupa usually took from a fortnight to three weeks to develop. The shortest period observed was ten days (temperature not unusually high).

The larva of this fly has already been described and figured*; but the mouth parts and stigmata need further elucidation to enable students to distinguish it from other allied species.

The great mouth hooks.—The anterior half of these organs is free, the remaining half subcutaneous; they are slightly falcate and bluntly pointed, the bases being emarginate, dorsally, with a clear nuclear space (? perforation) near the margin on the opposite side (fig. 12, *m.d. 2*). The *cephalo-pharyngeal sclerites* or processes (*c.s.*) for the first fifth of their entire length are of the same diameter as the mouth hooks, but at this point they suddenly dilate, forming a broad bifurcated plate; the lower or ventral half being broadest and longest with a deep posterior cleft. The *hypostomal sclerite* (*h.s.*), which lies between the anterior arms of the pharyngeal sclerites, is somewhat rectangular in shape, and resting upon it is a (? free) sclerite with three pairs of minute perforations; the anal angles of this plate carry a spine-like process.

External teeth.—There are three bilateral groups of these. The median pair (*c.t.*) are bidentate, and lie between the great mouth hooks; the anterior laterals (*p.s.*) form a distinct palmate group of nine stout teeth or spines; the posterior lateral groups (*p.s. 2*) are arranged in a similar way, but the outline is much more elongated

* First Interim Rep. of the Exped. to the Congo. Memoir XIII of the Liverpool School of Tropical Medicine.

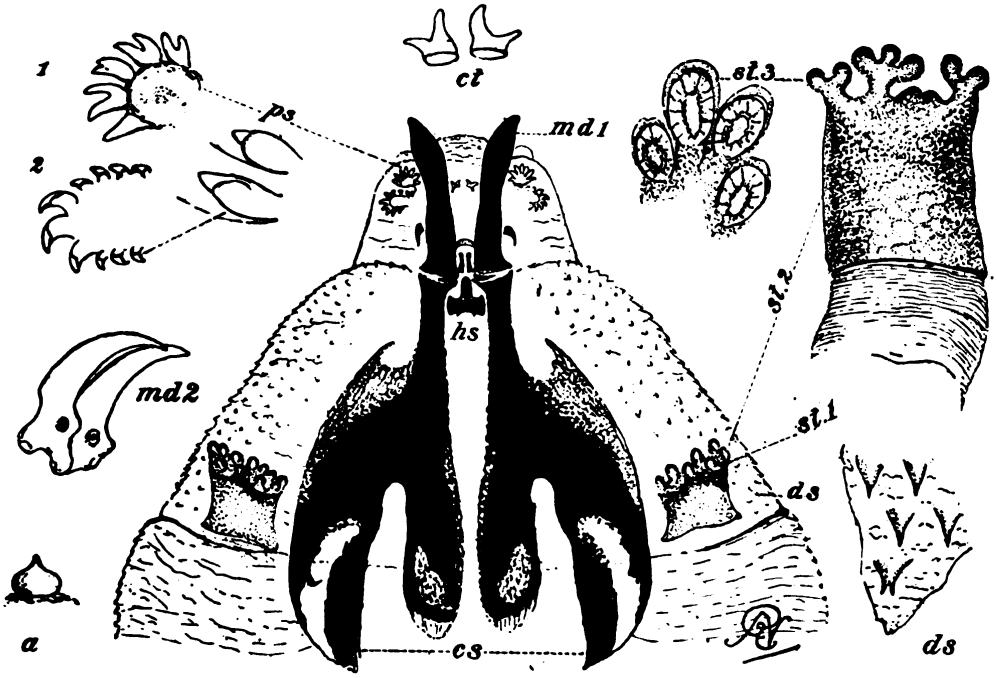


FIG 12.—*Auchmeromyia luteola* ("Congo Floor Maggot"). MOUTH PARTS, &c., OF LARVA:—*a*. tip of antenna: *md 1 and 2*. the great mouth hooks; *cs*. cephalo-pharyngeal sclerites; *hs*. hypostomal sclerite; *ct*. central or median teeth; *ps 1*. anterior-lateral palmate teeth; *ps 2*. posterior-lateral teeth; *st 1, 2, 3*. anterior or thoracic stigmata; *ds*. dermal spines. (All greatly enlarged.)

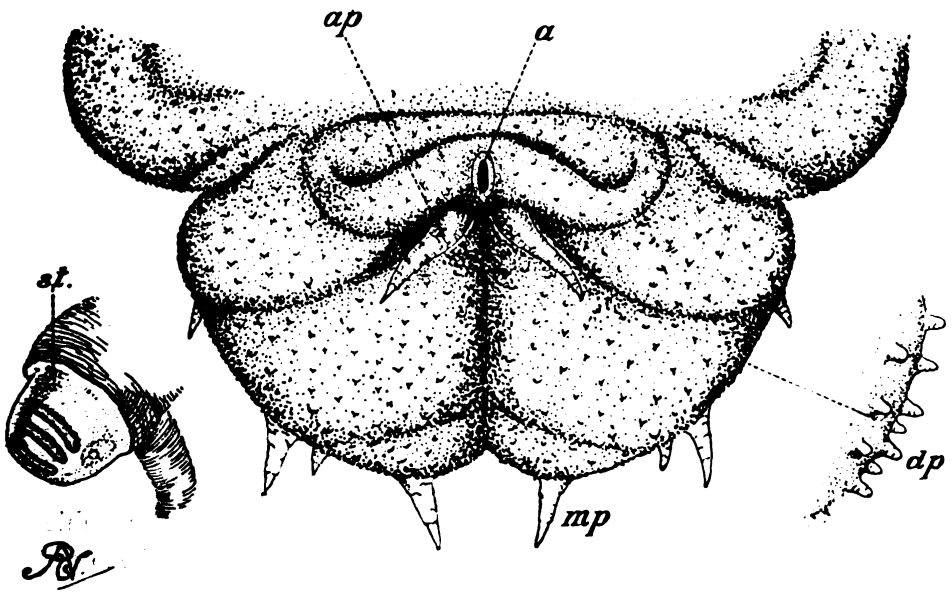


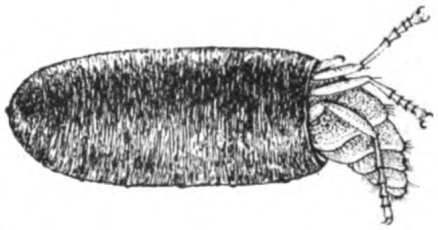
FIG 13.—*Auchmeromyia luteola*. ANAL SEGMENT OF LARVA:—*a*. anus; *ap*. anal papillae; *mp*. marginal papillae; *dp*. dorsal papillae; *st*. posterior stigmen. (All greatly enlarged.)

and the individual teeth or spines are distinctly curved upwards and inwards, and none of them are bidentate.

The *anterior* stigmata or spiracles (*st. 1, 2, 3*) are branched; each with ten to twelve relatively large orifices (*st. 3*). The *posterior* stigmata (fig. 13, *st.*) are large obconical projections of brown chitine; each presenting three darker transverse slits, partially closed by a fine transverse grating.

The posterior segment (fig. 13) bears along its margin four conspicuous bilateral papillae (*m.p.*), of which the median and third pairs are much the largest; the second and fourth pairs are much smaller; all are sharply attenuated and bear faint traces of segmentation.

On the method of the escape of the imago from the puparium.— The anterior pole of the puparium is invariably broken away by the imago on its escape, as is normally the case with other insects; but in four out of five specimens now preserved in the Museum of this



R. N. del

FIG. 14.—*Auchmeromyia luteola*. Showing escape of imago backwards from the puparium. × 7, about.

Institute the position of the imagines is reversed and the abdomen and legs are seen partly extruded from the anterior pole of the puparium (fig. 14), showing a very remarkable breach presentation. In all these cases, therefore, the imprisoned insect must have reversed its position in the puparium before the final instar was reached. In confirmation of this two pronymphs were found lying with their *heads against the posterior pole* of the puparium; as an explanation of this, however, one also finds that the *final larval stage is not reached until after the formation of the puparium:* the imprisoned larva retaining all the salient external characteristics of the previous stage, such as the mouth-hooks and papillae, &c. This being so, it is quite possible to conceive that the larva may reverse its position before it becomes

a propupa; though why it should do this is not clear. Neither is it possible with the limited material at hand to say whether the reversed position of the imago is abnormal or not. It is certain, however, that the pupa may also lie in its normal position with its head at the anterior pole of the puparium, as in dissecting out a number of puparia one imago was found in this position.

Thus we have a very remarkable anomaly; in the first instance the true larval stage is continued until after the formation of the puparium, and secondly, a large percentage of the flies escape backwards from the puparium.

Musca spp.

One species was taken on cattle at St. Louis, Senegal (May, 1903); another on cattle at Kasongo, and a third at Wanie Numbu. The specimens have, unfortunately, suffered somewhat from mould and are therefore unsuitable for descriptive purposes. Two of the species are apparently new to science.

A new Genus and Species of Blood-sucking Fly, allied to Musca

Several specimens of these interesting flies were caught on cattle and donkeys at St. Louis, Senegal, May, 1903, and also at Zambie on the Congo in September of the same year. All the specimens captured were forwarded to the British Museum in 1903 and 1904, and a series of five were returned to this Institute. Mr. Austen has given us to understand that he wishes to publish a description of this insect, and we await his publication* with interest.

Lyperosia minuta, Bezzi

Many specimens were caught on horses in a stable at Salikaine Creek, Gambia River.

Mr. E. E. Austen considers these as a new sub-species, having darker legs than the typical forms.

Lyperosia ? sp.

Specimens of a species allied to the above were caught on camels at St. Louis, Senegal, in May, 1903. We have not been able to identify these; but they are probably new.

* Mr. Austen has now (Jan. 18, 1907) very kindly promised to publish the diagnosis of this insect in an early number of this publication.

GENUS GLOSSINA (TSETSE FLIES)

NATIVE NAMES.—Only occasionally did the names given below accurately specify the insects for which they were used. For example, the word designating "tsetse fly" meant often only "a fly of medium size which bites," and therefore the same term was sometimes employed without distinction for *Glossina*, *Tabanus*, *Haematopota*, and even *Stomoxys*. In the same way the words given for "floor maggot" sometimes signified almost any maggot or even "a crawling thing which bites." *Glossina palpalis* is by far the commonest tsetse in the Congo. The names given for "tsetse fly" were obtained from natives by showing specimens of it.

The various languages are mentioned in the order in which the tribes speaking them were visited. When names are given from places off the route of the expedition the name of the informant is mentioned.

The names of native places and tribes are spelled according to the usage of the Congo Free State publications. The names of parasites are written in accordance with the rules proposed by the Royal Society for the orthography of geographical names.

DISTRICT OF THE LOWER CONGO.

Bacongo language.

Tsetse fly	-	*mavekwa* (*vekua*)
Maggot	-	*m'vidi* (at Matadi); *n'tungwa* (at Wathen, is a general name for all maggots)
Maggot fly	-	*kuluyanzi*

TSHUMBIRI.

Bateke language			Bobangi language		
Tsetse	-	*eyibi* (S)	-	*eyiyi* (S)	
		beyibi (P)	-	*biyiyi* (P)	

Note these words signify almost any Tabanus or tsetse; they are further distinguished by *etsii* = small, or *enene* = large.

Maggot	-	*mozeu* (S)	-	*libinzu* (S)
		mezeu (P)	-	*mabinzu* (P)

IREBU.

Kundu language			Lusakani language
Tsetse	*entune*	-	*vii*
Maggot	*n'kisu*	-	*kisu*
Tabanus	*ivovoko*		

chelifer (also bed bug)—*silibonga*

LULONGA.

Elehu language

Tsetse - *yiyi*
Maggots - *mabinzu*
Maggot fly (*Auchmeromyia luteola*) - *maboio*

UPOTO.

N'gombe language

Tsetse - *ipokupoku*
Maggot - *lutu*
Tabanus - *eholoholo*
Chrysops - *motuna*
Mosquito - *ngungu*

BUMBA.

Bangala language			Language (?) men from Loeka (Itembiri river)
Tsetse	*etuna*	-	*motoke*
Maggot	*kiso*	-	*n'kusu*

BASOKO.

Language (?)

Tsetse - *difu* or *lifugu* (seems to be special name)

YAKUSU.

Keli language

Tsetse - *makuku* (also signifies Tabanus)

STANLEY FALLS.

Bagenia language			Bakumu language
Tsetse	*kowowo*		
Maggot	*kalombo* or *bio* (?) (At Taritubu general name for all maggots is *kiyo*)	-	*kusu*
Tabanus	*dikuku*		

WANIE NUMBU.

Language (?)

Tsetse - *bungu* (*ngi* = any fly)
Mosquito - *m'bu*

People just below Wanie Numbu called *Glossina palpalis* '*kabobo*.'

KIRUNDU.

Swahili language (local dialect)

Tsetse - *chafua*

TO THE EAST OF THE CONGO RIVER AT LOWA.

Bujero language		Bakumu (?) or Babemo people	
Tsetse	- *otjapara*, said to be specific	-	*sumo* (S) *basumo* (P)

KUMBA.

Language (?)

Tsetse - *karboiboi*

KASONGO.

Swahili language (current trade language in the eastern part of the Congo Free State)		Bakusu language	
Tsetse	- *kibou kidogo*, i.e., a small biting fly	-	*mazanga*
Tabanus	- *kibou makubwa*, i.e., a large biting fly		
Stomoxys	- *ngi*, i.e., a fly		
Maggot	- *funza* or *funga*	-	*lukusu*

Bango Bango language

Maggot - *kivinya*

KATANGA DISTRICT.

Language (?) (Dr. Ascenso and others)

Tsetse fly - *kisembe* (*kasembe*) *kasembele*

CABINDA.

Basonge language

Tsetse - *kiboua* (*lundo* at Miambwe); same name is used for Haematopota

Maggot - *kifinya*

LUSAMBO.

Baluba and Bakuba people present

Maggot - *kikusu* (S)
bukusu (P)

Basongomeno language

Tsetse - *bohembe*

ANGOLA DISTRICT.

Language (?) (Bastian, native interpreter at Lusambo)

Tsetse - *dibubulu* (S)
mabubulu (P)

Maggot - *dindwe* (S)
mandwe (P)

LADO ENCLAVE.

Miza language (Commandant Charles Lemaire)

Tsetse - *ewe*

IN THE NORTH OF THE FREE STATE.

Mangbetu		Azande (natives of these tribes)
Maggot	- *ngousu*	- *agbiti*

AT KOUSSOUR ON THE SCHARI RIVER NEAR LAKE TCHAD.

Maggot - *tadi*

Maggot fly - *acou*

This information was given by an intelligent lad in the service of the expedition led by Monsieur Chevalier.

Glossina palpalis, Rob.—Desv.

(Pl. iii, figs. 7-9)

Larva.—Orange-yellow; hood or anal segment black. Form somewhat cylindrical. Segments very pronounced; transversely wrinkled, and finely tesselated, the individual tesserae being flat and

shiny. Mouth hooks minute, contiguous and black. Anal segment or "hood" with the tumid lips deeply divided and widely separated.

When fully matured the larva almost completely fills the abdominal cavity of the parent, and lies with its posterior extremity ("black hood") towards the vaginal opening. A female which died while in the act of parturition has been preserved with the anterior extremity of the larva still attached to the vaginal orifice. This interesting phase in the bionomics of this tsetse fly is illustrated on Pl. iii, fig. 7.

Length 4·50 mm.; width 1·75 mm.

These measurements were taken from larvae laid by captive flies among whom abortion is frequent. These larvae may, therefore, not be quite fully matured.

Pupa.—This agrees so well with the description given by Austen* that it is necessary only to call attention to the following details:—Colour, in a bright light, dull steel-blue, but traceable only in perfectly clean specimens. The tumid lips are divided to their bases and widely separated.

Length 5 to 5·75 mm.; width 3 mm.

Localities:—*Glossina palpalis* is considered to be certainly present at the localities mentioned in the following list. Places at which specimens were obtained by the expedition are, as usual, simply mentioned. If the specimens were donated by friends their names and the date are given.

The names of several villages, from which no specimens have been obtained, appear on the list. *G. palpalis* is believed to have been present at them because of the proximity of places in which it has been seen and because of the evidence asserting its presence. The nature of this evidence is in every instance mentioned. It is indicated whether persons unacquainted with the name of the fly were shown specimens and *recognised* it as being present, or whether it was *reported* as present by those knowing its name. As a rule such evidence cannot be accepted as indicating more than the presence of a small tsetse (not *G. fusca* or *longipennis*). A second and supplementary list has therefore been prepared of places from whose neighbourhood specimens have not been received, at which "tsetse flies" have been reported to exist. In Map II the markings indicating the

* Monograph of the Tsetse Flies. Brit. Mus. Nat. Hist., p. 26 (1903).

distribution of *Glossina palpalis* are based upon the first list; those showing the distribution of undifferentiated species of "tsetse flies," partially depend upon the second list.

Banana (Oct., Dr. Etienne); Shiloango, Lukula and Lubengi Rivers (reported, June, 1905, de Laval); Banza Manteka (reported); Lufu River (reported, Rev. Morgan); Tumba; Wathen; Kuzu; Kisantu; Sabuka; Leopoldville (Nov., 1905, to July, 1905); Brazzaville; Bamu Island (Jan.-Feb.); Lisha (April, June, July, 1905); M'Swata; Kwamouth; Kitoto; Tshumbiri (seen ten miles inland, said to exist along all small streams of neighbourhood); Bolobo (recognised as present); Yumbi; Lukolela; Irebu; Bikoro (reported by natives and Rev. Clark); Bolengi; Coquilhatville; "All along Lulonga River to Baringa" (July, Dr. Angela); Ikelemba River (Nov., Major Malfeyt); Eala; Bamamia; "All along overland route from Boyembe to Lulonga" (July, reported, Rev. Gilchrist); Lulonga; Monsembe; Nouvelle Anvers; Bosesera (reported, Jan., Chef de Poste); Mobeka; Bokanga; Lisala, Bwela, Bosogodo (May, reported, Rev. K. Smith); "Along Congo from Lisala to Bumba and up the Itimbiri River" (reported, Dr. De Valkeneer); Bumba; Botsali; Yambinga; Bopamba; Basoko; Yalembe; Isangi; Yarbumbo (recognised by natives); Yandonge (recognised by natives); Yakusu (recognised by natives and Rev. Milman); Stanley Falls (recognised by natives); Katanga (recognised by natives); Batikalela; Wanie Bakula; Kewe; Wanie Numbu; Kisui; Ponthierville; Kirundu; Lalowa River; Utikakadjia; Lulindi River; Kumba; Kasuku; Maboka; Lokandu; Ukungwa; Kamimbi; Sendwe; Makula; Mfunkiva; Kibombo; Kundu River (Dec., reported, Commandant Verdick); Nyangwe; Salt Springs (Oct., reported, Monsieur Sappen); Kasongo (Oct. to May); East Bank of Lake Albert (April, Commandant Engh); Lado Enclave, 6° 33′ N., 29° 58′ E. (March, Commandant Lemaire); "*Glossina palpalis* is everywhere present in Uele, even in the smallest rivers" (Feb., 1906, reported, Dr. Hosselet); Kalombe (May, recognised by natives); Muadi River; Maomedi; Tshofa; Miambwe (recognised as present by natives); Kabinda; Kiambi; Katanga (Aug., Major Malfeyt); Pania Mutombo; Batampas; Lusambo (none at post itself, many along Lubi River, just opposite, and up and down Sankuru River); Lubefu; Basongo; Luano (Kwilu River, July, Major Malfeyt). There were but few *G. palpalis*

among the grass-covered islands of the Lower Kasai. They were, however, numerous in every patch of forest. In the Gambia (Sept., 1902, to April, 1903) *Glossina palpalis* was everywhere present along the river and its tributaries from its mouth to a point some fifty miles above Fatotenda.

Circumstances of capture: -On reviewing this list and comparing it with Maps I and II, it is apparent that *G. palpalis* is probably present in almost every part of the Congo Free State.

Tsetses were usually only found near water. If there be a fringe of forest or brush, perhaps 200 yards in breadth, along the water's edge, more flies will be seen by gently paddling in a canoe along the bank or by walking on the land side just at the edge of the belt of brush than by even a prolonged stay in the forest itself. The collection of water need not be large. *G. palpalis* and *fusca* have been caught along very small forest streams.

Their numbers were found to vary greatly in different localities. For example, along the Congo at Sabuka, near Coquilhatville, near Sendwe and near Kasongo, on the Lurimbi River at Tshofa, and at Batampas and Lubefu on the Kasai it was, as at many other places, almost impossible to avoid being bitten; but at Zambie, about the outskirts of Boma and Matadi, at Bolobo, Lisala, Romée and Stanley Falls, all places at which we believe *Glossina palpalis* to be present, not a single fly could be found. Careful search was made during our stay in each of these places, both by ourselves and by our fly-catching boys; it may be that we were unfortunate in visiting some of them on days when the flies were not visible, since it has been repeatedly noticed that the number of tsetse flies present in any locality may vary considerably from day to day without apparent cause.

On October 15th one member of our expedition left Maboka on the Upper Congo for Kaya. No tsetse flies were seen during the first half of the journey, and only one during the latter half. Ten days later the remaining members of the expedition travelled over the same piece of river and found large numbers of flies along the whole route. (See also Memoir XIII, Liverpool School of Tropical Medicine, page 106).

Again the flies may have been absent from some of the places named because they were comparatively free from brush along their water fronts.

It seems certain that the number of tsetses in the immediate neighbourhood of a post of any size may be considerably reduced if

all thick underbrush and rank grass, either near the water or a few hundred yards inland, be carefully removed.*

The station of Irebu, a military training post on the middle Congo, is beautifully kept. We remained there for three days and saw no tsetses, although there were many only a few hundred yards further up or down the river. A little over 1,000 yards outside the station was a trading factory placed in the middle of a patch of rank grass. *Glossina palpalis* existed there. Besides searching for tsetses on the station ourselves we sent out five boys trained to catch them, together with 60 soldiers. Between them all only a dozen specimens were taken. All were caught in a patch of rank grass, about 350 yards from the river, that had not been cut for two or three months. There were many scores of acres of cultivated land on the station, mostly consisting of well-kept, but shady, cocoa and coffee plantations. In them not one tsetse was found.

As Leopoldville developes and the surrounding brush is kept down tsetses become fewer, and we are told they are not nearly so numerous within the Protestant mission station as they formerly were. During our stay in Leopoldville *Glossina palpalis* was much more often caught in the house of the Rev. Mr. Morgan than in the building occupied by our expedition. The mission house was surrounded by a grove of palm trees which extended uninterruptedly to the river's edge, while the house of the expedition stood in an open piece of ground.

The experimental farm of the Government of the Congo Free State at Eala possibly may be considered to be an exception to this rule.

This farm is comparatively free from undergrowth. It is extensive and well kept, yet *Glossina palpalis* is quite frequently seen on it. There is a herd of 43 cattle and a fair amount of other live stock on the farm. It is believed that the flies are attracted by them.

It often happened, however, that no, or extremely few, tsetses were found, even after a careful search, in places where everything, shade, shelter and water, apparently favoured their presence.

Near Yambinga a search (of several hours), made on a bright day, along the thickly wooded banks of the main river of a small stream failed to find *Glossina palpalis*, although they were present in the near neighbourhood.

At Yakusu several hours in the middle of a fine dry day were spent in a canoe paddling along the banks of the river and about wooded islands. Tsetse flies were not seen.

The late Rev. W. Holman Bently told us that at a place called Vela in the Lower Congo *G. palpalis* was absent. The natives noticed this and decided that they were suffering from a want of the beneficent blood-letting enjoyed by their neighbours who lived in places where the fly existed. Men were therefore sent out to catch tsetses, bring them back and let them loose at Vela!

Behind the soldiers' lines at Lusambo runs a brush-covered stream which never

* d'Aguiar. La maladie du sommeil et la tse-tse à Novo Redondo. Reports of the Fifteenth International Congress of Medicine at Lisbon, Fasicule 2, page 294. It is stated in this publication that *Glossina palpalis* is not seen in well-kept plantations nor at a higher altitude than 400 metres above the sea. This latter assertion is negatived by an observation of Commandant Charles Lemaire, who sent us specimens of the fly from the banks of the Ialo River (6° 33′ North; 29° 58′ East) caught at an altitude of 530 metres, and also by Lieutenant Brohez, who caught *Glossina* at an altitude of 1,600 metres, in places where the morning temperature went down to 0° C. (*loc. cit.*, *vide infra*).

goes dry, and in the wet season measures from ten to twenty yards in breadth. One could not imagine a place better-suited for tsetses; yet three competent observers (Dr. Broden, Dr. Polidori and J. L. T.) during a period of several months saw none there, although they were looked for many times. The observation might be explained by the fact that the stream is used as a latrine, and in hot weather reeks with effluvia. If this explanation be accepted it seems strange that *G. palpalis* has twice been seen in latrine sheds.

Tsetse flies will sometimes follow persons or animals for a considerable distance.

In the Lower Congo the belief that *G. palpalis* follows pigs is very common. We have not confirmed the observation. We are told that in the neighbourhood of Leopoldville *G. palpalis* was called by some of the natives "the pig fly" (Dr. Broden).

At Kilobala, a cattle station in the Lower Congo, it is definitely asserted that tsetses were formerly absent. Pigs were introduced, and with them appeared the flies! (?)

In the Gambia we lived for a time in a house placed almost a mile from the brackish water of a mangrove swamp where there were a good many *G. palpalis* Flies were twice carried into the house by persons who had just come over the road through the swamp. These were the only occasions that tsetses were seen about these buildings during a residence of three months.

At Lokandu on the Upper Congo, no tsetses had been seen about the house we used as a laboratory. Two tame antelopes were brought to be examined; two or three *G. palpalis* followed them.

Apart from these observations no information was gathered in the Congo to support the idea that tsetse flies are dependant on large game. On the contrary, many *G. palpalis* were seen in localities where there was exceedingly little game of any sort.

Apparently because of this habit tsetse flies were occasionally not seen at places where they were ordinarily present.

When the expedition reached the Lomami River, during the journey overland from Kasongo to Pania Mutombo, it employed some 300 porters. These natives reached the river first, and not a single tsetse was seen by the European members of the expedition during the hour and a half at midday spent in ferrying the caravan across the river. The flies were later frequently seen in large numbers at this spot at all times of the day.

It is probably for this reason that tsetse flies were not found at several of the small streams crossed near Kalombe, Dibwe, Miambwe and Lukola during the overland journey of the expedition. Search was usually made at the fords and bridges after a considerable number of porters had passed or while they were still present. The flies were not found in several instances, although the natives asserted their presence and although human trypanosomiasis existed locally. A search of one or two hours discovered tsetses at a couple of streams where they had at first seemed to be absent. At other water-courses a search during a like period was without result, and it seems possible therefore that *G. palpalis* only occurs in small numbers at the following places where human trypanosomiasis is frequent—the

percentage of resident natives having enlarged cervical glands without apparent cause is indicated—Kalombe (17 per cent.), Dibwe (18 per cent.), Miambwe (7 per cent.), Lokula (6 per cent.).

A proportion of the infected persons probably contracted their disease elsewhere. These villages have a considerable immigrant population ; they supply many carriers and a large proportion of their people are continually absent, collecting rubber, in the forest near rivers where tsetse flies are said to exist.

Had the flies been present in as large numbers as, for instance, at Kamimbi, it is quite inconceivable that the passage of even 300 men should have enticed them all away from their usual haunts.

The occasional presence of *G. palpalis* in places at a considerable distance from any collection of water may perhaps be explained by their habit of following animals.

In the Gambia, and again in the Congo, a single *G. palpalis* (?) has been seen on a dry plateau a couple of hundred feet above and about a mile distant from the nearest stream.

As would be surmised from what has already been said, *Glossina palpalis* is able to fly quickly and for comparatively long distances.

They are said to occasionally fly into the railway carriages as they pass near Palabala in the Lower Congo. They are frequently seen on steamers or canoes in mid-river, and distant 300 to 500 yards from either bank. On entering a moving vehicle the flies seek sheltered spots, and it is under the sunshade of the canoe or in corners of a cabin sheltered from the wind that they take refuge.

On one occasion in the Gambia a *G. palpalis* (?) persistently followed one of us (J. E. D.), who was riding rapidly on a bicycle, for several hundred yards.

At Tshofa, *G. palpalis* was frequently seen on the verandah of our house, although it was placed on high ground and at least two hundred feet above the river, which was about half a mile away.

Although *G. palpalis* has such extensive powers of flight, it seems to be very local in its habits. As has often been observed, not a single fly may be seen at 100 yards from a river although its banks swarm with them.

At M'Swata, however, *G. palpalis* was regularly seen along a little-frequented path on high ground and in open park country at a distance of 500 yards from the water and of 250 yards from the edge of the fringe of brush bordering the river. In partial explanation it should be stated that there was a good deal of game in the neighbourhood.

It has been often noticed that *G. palpalis* is most numerous at, and seems to lie in wait by, fords or frequented paths. Its habits have seemed to be more or less regular.

On one occasion, while resting by the side of a path, a *G. palpalis* was noticed to return on three occasions to the same spot on the sheltered under surface of a grass stalk after having been driven off by a native porter whom it was trying to bite.

Tsetse flies were practically never seen during rain and wind. They were most conspicuous and seemed most vivacious on bright,

warm days. In the cool of the early morning or on chilly days they were sluggish and were certainly much more easily captured; under these conditions tsetse flies have often been caught with the hand, ordinarily almost an impossibility.

Glossina palpalis is often seen in native villages and in the houses of Europeans living within a few yards of the river's edge.

The Rev. Mr. Morgan asserted that they most often entered his house on warm days. At Monsembe, on a cool morning (minimum temperature 71° F.) *G. palpalis*, so sluggish that they were caught between finger and thumb, were seen in the house of the resident European.

Field notes on the Bionomics of tsetse flies.—These notes are based on observations made on the captive flies, used during our attempts to transmit trypanosomes by their bites, and on flies seen in freedom during our stay in Africa.

When a tsetse prepares to feed* it spreads its legs, particularly the first pair, and so brings its whole body nearer its host; thus, as it were, "settling down to business." The palps are retracted until they form an angle of about 135 degrees with the bared proboscis There is occasionally a moment or two of hesitation before the proboscis is inserted. Ordinarily there is none and in a second or two the whole proboscis is buried to its bulb.

The proboscis seems very powerful and the flies easily pierce socks and ordinary white drill or duck clothes. They were, however, on more than one occasion seen to be quite unable to penetrate the tightly woven texture of a pair of riding breeches made of a patented cloth.

The penetration of the skin by their proboscis sometimes causes considerable pain. Occasionally it is unfelt.

Specimens of *G. palpalis* have frequently been watched filling themselves with blood from the backs and legs of unconscious paddlers.

While coming down the Kasai river, a small deck cabin was fitted up as a laboratory. Doors and windows were guarded by wire gauze, but through carelessness three tsetses had been allowed to enter the room. When they were found all were gorged with blood. The occupant (J. L. T.) had felt no bite. The weals, evidence that he had been bitten, soon appeared.

The proboscis, ordinarily completely inserted at the commencement of feeding, is usually withdrawn for a short distance just before the fly commences to visibly fill with blood. Often blood is not obtained on the first trial, and either the proboscis is withdrawn altogether and operations are commenced in a new place, or it is partially withdrawn to be re-inserted in, apparently, a new direction. The following observations will perhaps explain the way in which this

* *G. palpalis* was watched with a pocket-lens while feeding.

may be done. It seems that *G. palpalis* flexes its proboscis in two distinct ways. The first, the more usual, is a sharp bending of the labella. Flies who had just fed were often seen, while preening themselves, to sharply flex their labella so that they stood out, almost at right angles, from the palps. The second movement was only seen in flies from whom the palps had been removed preparatory to dissection. In these the whole proboscis bowed until it described a well-marked curve directed ventrally. The second movement is probably due to the contraction of the powerful "tendons" lying in the labium.*

G. palpalis does not fill with blood so quickly as is usually imagined. The length of time between the insertion of the proboscis at the commencement of feeding and its withdrawal was noted in 25 flies carefully fed on an experimental animal. The greatest care was taken to avoid disturbing them. The longest fed for eleven minutes, the shortest for 1½ minutes, while the average was slightly over three minutes.

The extraordinary way in which tsetses distend themselves with blood is well known.

A female *G. fusca* was watched steadily filling itself with blood for six minutes. It was stopped feeding then lest it should burst itself. Its abdomen was so distended that there was a space of almost a millimetre between each of the dark coloured chitin plates on the dorsal surface of its abdomen.

In about a minute after feeding commences a drop of yellowish or brownish, opaque, but liquid faeces is extruded and a minute later—while still feeding—clear serum rolls, drop by drop, from the anus.

Sometimes tsetses who were plainly in need of food absolutely refused to attempt to feed. Others frantically probed animal after animal and failed to get blood. It has occasionally been thought that in captivity male flies did not seem to be so eager for blood as were the females.

In the cool of the early morning or evening *G. palpalis* is not nearly so voracious as at mid-day.

One is, however, occasionally surprised, as at Lubefu on the Kasai, in finding them active and biting freely early on a cold damp morning, or as at Kasongo and Lukolela in seeing them feed until dusk and fly actively about after dark. *G. palpalis* has twice been caught about the lamps at night.

These flies sometimes seem to prefer natives to Europeans, and dark to light coloured clothes. This first point is frequently commented on by Europeans travelling in canoes. They see that the native paddlers are worried by the flies while they are themselves often comparatively unmolested.

* Memoir XVIII of the Liverpool School of Tropical Medicine, page 53.

One day while coming down the Kasai the Captain, finding it cold, put on a blue cloth jacket. Many more tsetses were noted to settle on his coat than on the white duck suits of the European passengers and of the negro steersman.

The flies seem to find their hosts rather by sight than by smell, since in a given locality a moving person is usually more troubled by them than is one who sits still.

The persistency with which *Glossina palpalis* resumes its attacks despite slapping and brushing is very characteristic. As has already been stated, its flight is ordinarily rapid and darting. It is usually accompanied by a peculiar and characteristic buzzing. This fly can nevertheless, if it wishes, fly as noiselessly as an *Anopheles*. A fully gorged fly is exceedingly heavy on the wing, and usually flies only a yard or two before taking refuge in some quiet corner, where it quickly lightens itself by the rapid extrusion *per anum* of large quantities of colourless serum. Immediately after feeding the fly goes through an elaborate toilet and carefully cleans its proboscis. Sometimes just after feeding a drop of blood has been seen adhering to the outside of the proboscis at about its middle.

If a tsetse (*G. fusca* or *G. palpalis*) be held by its wings, or otherwise irritated, it can often be made to exude a tiny drop of clear fluid from the tip of its proboscis. Koch has made a similar observation.*

It has often been said that tsetses are frequently found in association with a particular tree or shrub. In the Congo two such plants were pointed out to us. One was a peculiar sort of palm, the other a variety of mimosa. Both grew on low-lying land along the river bank, and therefore were always seen where tsetses existed. Needless to say, the flies have been frequently seen in places where both were absent.

Certain reports to the expedition as well as the fact that *Glossina* was found to be absent from places apparently well suited to it have raised the question whether the numbers of tsetse flies present in a given locality may not vary very greatly through seasonal or other influences. No satisfactory answer has been obtained. The following observations are, however, interesting in this connection.

At Matadi and elsewhere in the Congo, as also in the Gambia, we were told that tsetse flnes (*G. palpalis*) were much more numerous in the wet season. Lieutenant Brohez† asserts the contrary, and states that in Katanga they are more numerous and bite more fiercely in the dry season (*G. morsitans*, *G. longipalpis*, *G. palpalis* present).

* Koch, Vorlaufige mitteilungen über die Ergebnisse einer Forschungsreise nach ostafrika ; Deutsche medizinische wochenschrift, No. 47. Nov. 23, 1905.

† *loc. cit. infra.*

In April, just at the end of the dry season, a camp was made on the banks of the Upper Gambia river. *Glossina palpalis* was very numerous, but, strange to say, caused but little trouble ; there was a great deal of game in the neighbourhood.

It has been reported that certain localities were infested with tsetses which at an earlier period were free from them. These statements have not been substantiated, and they seem sometimes to have resulted from insufficient observation. The possibility of their accuracy must not, however, be forgotten, especially since it is frequently asserted that the area over which *Glossina* exists is widening.

Observations on captive Glossina palpalis.—During our attempts, in the Gambia,* to transmit trypanosomes by the bites of tsetse flies, we found that they would not feed well in small cages and died in the course of two to four weeks. It was thought that this might have been one of the reasons why our Gambian experiments had been unsuccessful, and when we recommenced our experiments in the Congo we determined to reproduce, as nearly as possible, the natural surroundings of the fly by keeping them out of doors in larger cages containing water and growing grass. Cubical cages of wire gauze measuring 18 inches along each side were employed. In each cage was placed from 20 to 70 flies. These larger cages were kept in a large mosquito house, measuring 13 by 8 by 9 feet, made of wire gauze. It was placed in a thick clump of bamboos, distant about 100 yards from the river. The flies were sheltered by the bamboos from the mid-day sun and, on two sides, from strong winds. Food was supplied to them by experimental animals, guinea-pigs and rats, which were immobilised and placed in the cages. *Glossina palpalis* certainly lived better under these conditions than they had done in smaller cages in the Gambia. They frequently lived for over 30 days ; 43 and 59 days were the longest lives noticed. In addition they seemed not to require food so often as had been necessary in the Gambia, and they were frequently left for 48 hours without an opportunity of feeding. This may be explained in part by their being out of doors and more exposed to cold and damp. Under such circumstances tsetses are certainly more sluggish in their movements.

The disadvantages of this method of keeping flies were however great. It was almost impossible in such large cages to count exactly the number of flies which had fed or to estimate the number present. They seemed to seek concealment and hid themselves in the smallest crevices. It was difficult to feed flies kept in this way on monkeys and the cages were too large to be easily transported.

* Memoir XI, Liverpool School of Tropical Medicine.

In our experiments at Kasongo we therefore once more returned to the smaller cages. During our stay at Kasongo (Dec., 1904, to April, 1905) the average number of flies caught per day by each of the twelve members of our fly-catching gang was only about thirteen.* In spite of rewards for anything over twenty flies and punishment for anything under ten, the lads and men employed refused to exert themselves. Had they wished, each of them could have easily brought in thirty flies daily. Although it has been noticed that tsetses, *G. palpalis*, are far more "lively" at midday than on cool mornings, and although, on one or two occasions (at Leopoldville), particularly large numbers of flies have been brought in on warm days, the temperature seems to have had no marked influence on the number of flies caught by our boys at Kasongo. Most of the flies were caught on a small forest and brush-covered island about two miles down-stream from Kasongo. A few were caught along the river banks above and below Kasongo. All were taken between 7.30 a.m. and 4.30 p.m. The great majority of the flies captured were *Glossina palpalis*; among the 1,053 flies actually examined with a lens, only one *G. pallidipes* was seen. This was the only fly of this species we saw in the Congo. About a score in all of *G. fusca* were taken at Kasongo. There was no preponderance of either sex among the flies caught.

The method employed in taking the flies was as follows. Each negro was provided with a short-handled, deeply-bagged "butterfly" net. When the tsetse, in attempting to bite, alighted on the natives' bare legs the net was whisked by and the fly was usually caught. It was then transferred to a dry, dark-coloured bottle; they were thus protected from sun and wind and lived much better than in small wire gauze cages. Flies collected in white glass bottles were invariably killed by the sun in a very few hours, while on several occasions strong winds have seemed to be very destructive to flies in cages. On arriving at the laboratory the flies were transferred to three-sided wire gauze cages from six to ten inches in length, with sides four inches in width. The bottom was made of wire gauze and the top was closed with white cotton cloth and a purse string gathering. As many as 70 flies were occasionally placed in one of

* The average number of flies caught by the fly-catching "gang" at Leopoldville (Nov., 1904, to Jan., 1905) was only slightly better than this.

the largest of these cages. The usual number was about 30, often less. No water was placed in the cages and they were simply hung in a sheltered, shaded corner of the laboratory, out of the way of ants; on one occasion several hundred flies were practically all killed in one night by small red ants.

Under such conditions the mortality amongst the flies, though well fed, was very great. Especially was it so during the first few days in captivity before they seemed to have realised that they were caged. They continually dashed themselves against the wire gauze and in a very few hours had frayed their wings to mere stumps or occasionally had even lost a leg or two. As they became accustomed to their surroundings they became much quieter, barely moving for hours together, and the death-rate diminished.

Thus, about 32 per cent. of the flies died during the first 24 hours, 11 per cent. during the eleventh 24 hours, but only 9 per cent. during the seventeenth.

One fly, from a batch of 19, lived under these conditions for 38 days. Many lived for from 20 to 28 days. It has been thought that perhaps the males live longest in captivity. The frequency with which gravid female flies aborted soon after entering the cages was very noticeable. Also remarkable was the frequency with which pairs were seen in copulation and the length of time they remained *in coitu*. Frequently a dead fly of either sex was seen linked to one of the opposite by the powerful male claspers.

Only three of the ten apparently mature pupae obtained by us ever produced imagines (in about a month's (?) time, not longer). Three of the pupa were of *Glossina fusca*, none of them hatched. One female *G. fusca*, kept in captivity for over three weeks, deposited an apparently mature larva on March 28th, and another in the afternoon of April 1st. When found, very shortly after extrusion, the larva was white. An hour later the pupa had become a light brown. At the end of the second hour, it was much darker; after $4\frac{1}{2}$ hours the pupa was almost black, but it was noted that the under side seemed less dark than the upper side exposed to the light.

So long as the flies were fed once in twenty-four hours the frequency of feedings and the numbers of trypanosomes, often enormous, ingested with the blood seems to have had no effect on the length of their lives.

If they were left without an opportunity to feed for much more than 24 hours they died very quickly, about 91 per cent. in 37 hours. Neither sex seemed particularly resistent.

To feed the flies the cage was simply pressed against an infected person or animal, and so long as the victim kept fairly still the flies

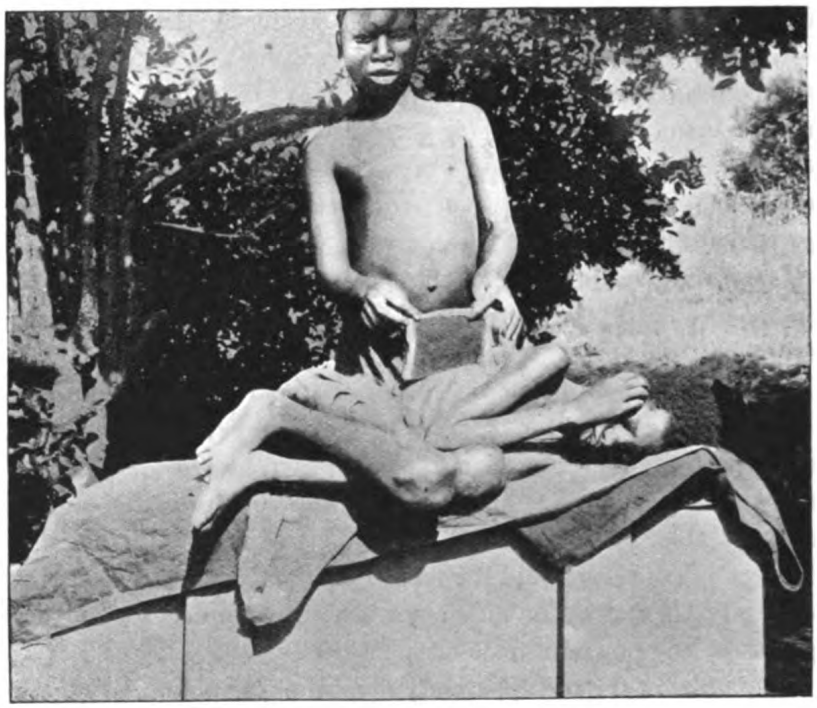


FIG. 15. METHOD OF FEEDING TSETSE FLIES.

fed without difficulty. They showed no preference for human blood, and practically the same percentage of flies fed on whatever animal (man, monkey, guinea-pig, &c.) was offered. On one occasion they were persuaded to suck blood from a frog.

It is calculated from observations on several thousand flies that on an average 80 per cent. of our flies fed when the opportunity offered. In special cases, when great care was used to keep animal and cage perfectly still and the feeding was persisted in for some hours, much larger percentages were obtained—frequently 100 per cent. Flies which had only partially filled themselves could often be persuaded to at once resume their meal on a second animal. It was practically

impossible to get flies which had been allowed to gorge themselves to again feed within from three to five hours' time, although they would do so readily eight hours after a first feed.

It has already been stated that *G. palpalis* may buzz loudly while flying. When at rest, often just after feeding, it sometimes produces a shrill, high-pitched note. A wingless fly was watched with a pocket-lens while sounding in this way. The tips of its halteres were seen to be depressed downwards and forwards, but the mechanism by which the note was produced was not ascertained.

Glossina palpalis, var. *wellmani*, Austen

Localities:—On the Congo River, between Nyangwe and Kasongo; on the Gambia River, near its mouth, at Oyster Creek.

Circumstances of capture:—So far as was observed the habits of these flies were quite similar to those of *Glossina palpalis*.

Glossina morsitans, Westwood

Localities:—Specimens have been received, through Monsieur Brohez, only from the Katanga district. The exact locality and the circumstances under which they were caught are not certain, but they were probably taken along the Luvua River near Pueto. Dr. Hosselet definitely reports that this fly is everywhere present in the zone of Uele, but we have seen no specimens.

Glossina fusca, Walker

Larva (dissected from body of parent but fully matured).—This consists of twelve strongly defined segments; surface finely tesselated, the individual tesserae being circular, flat, and shiny. Form, rather short ovate. Mouth-hooks relatively large, black, rather widely separated, but not porrected. Anal segment or "black hood" very coarsely rugose, the space between the converged points of the tumid lips one-third the diameter of one lip. Colour white, with a trace of lemon-yellow; hood or anal segment black.

Length 5·50 to 6 mm.; width 3 to 4 mm.

Pupa much more tumid and ovate than that of *G. palpalis*, especially so posteriorly. The tumid lips are also more depressed, and being continuous at their bases and only slightly divided posteriorly form a deep siphon-like tube or crateriform process, so

deep that it is impossible to see the stigmata which lie in the interior. Colour, dull steel-black, without any trace of the dull steel-blue seen in the puparium of *G. palpalis*.

Length 7·50 to 8 mm.; width 4 to 4·50 mm.

Localities:—Leopoldville (Dec.; reported, all the year round); Tshumbiri; Yumbie (Dr. Rodhain); Lukolela (Rev. Whitehead); Bolengi (Dr. Dye); Coquilhatville (Dr. Mordighlia); Nouvelle Anvers; Bwela (reported, Rev. Kenred Smith (*longipennis* ?)); Aruwimi River; Lulu River (reported, Commandant Lund (*longipennis* ?)); near Lalowa; near Utikakadja; Sendwe; Mfunkiva; Kasongo (Oct.-May).

Circumstances of capture:—This fly occurs in very much smaller numbers than *G. palpalis* and it is the rarest thing for more than one to be seen at a time. Most of our specimens were caught in the bush or from canoes paddled slowly along the edge of a wooded stream. None were seen to fly on board the steamer used by the expedition on their journey from Leopoldville to Stanley Falls, although a dead *Glossina fusca* was found in one of the cabins. *Glossina fusca* has frequently been observed in the Congo to be more or less nocturnal in its habits.*

A buffalo was shot. Part of the carcase was sent by canoe to camp, where it arrived about an hour after nightfall. By the light of a lamp a large *fusca* was caught trying to feed—there was no blood in its stomach—on the buffalo's neck. A night or two later a pair of *G. fusca* were caught, *in coitu*, about the lighted lamp in a tent. On two occasions specimens have been caught after nightfall in the illuminated tents of the expedition.

One or two observations on the bionomics of this fly have been included in the notes on captive *Glossina palpalis*.

Glossina pallidipes, Austen

A single specimen of this fly was obtained at Kasongo. No others were seen, although 1,053 tsetse flies, caught for transmission experiments, were carefully examined.

Glossina longipalpis, Wiedemann

Localities:—This fly has only been sent to us from the Katanga district. Two lots of specimens were received. One reached us through Major Malfeyt, the other was sent by a local official from the Upper Luapula.

* Memoir XIII, Liverpool School of Tropical Medicine.

Field notes:—It is asserted locally that these flies are more persistent and feed more viciously than *Glossina palpalis*.

Glossina maculata, n. sp. (Newstead)

(Pl. iii, figs. 5, 6)

General appearance.—Very dark brown; posterior surface of head cinerous, spotted with black; thorax dark brown and faintly cinerous in places, with elongated transverse black spots; pleurae, coaxe and femora cinerous with conspicuous black spots; first and second segments of hind tarsi dark. (The remaining segments wanting.)

FEMALE.—*Head* with the frontal stripe rich ochreous brown; frontal margins and occellar spot bright yellowish-white to pale ochreous; ocelli black; posterior surface of head cinerous *with large well-defined irregular black spots*. *Antennae* dull red-brown in front, sides with a cinerous surface; arista dull reddish-brown. *Palpi* blackish above, paler beneath. Bulb of *proboscis* dark castaneous, shining. *Thorax* dark brown and cinerous with numerous irregular elongated black spots placed transversely; anterior angles pale brown. The scutellum spotted like the thorax, margin pale brown; pleurae cinerous and pale brown, with numerous, irregular, and more or less confluent, black spots. *Abdomen* very dark brown; the narrow basal segment cinerous, with numerous black spots; the broad second segment with the large median area, the anterior angle, and broad hind margin, cinerous, with black spots; the remaining segments, with the exception of the last, with a narrow well-defined median stripe, and hind margins narrowly cinerous; lateral margins of segments three to five with a cinerous triangular patch; the hind margins with an occasional, more or less obscure, black spot, with the exception of the sixth, which has a regular series of ten or eleven on the grey cinerous band; last segment darker than the rest with a lateral greyish spot in the centre of which is a small black one. *Legs:* All the coxae cinerous and pale reddish-brown, with numerous more or less confluent black spots; anterior and mid femora cinerous basally and apically bright red-brown, cinerous area with numerous confluent black spots; hind femora with the basal third and apex pale red-brown, the rest faintly cinerous with a few large, faint, dull brown blotches; anterior and hind tibiae pale brown, mid tibiae slightly darker with, in some lights, faint indications of dark spots; anterior

tarsi and first segment of the mid pale brown; first and second segment of hind tarsi dark brown. (The remaining segments of the mid and hind tarsi are unfortunately wanting.) *Wings* uniformly brown.

Length,* exclusive of proboscis, 9 mm.; length of wing 9 mm.

A single specimen of this fly was sent to us by the Rev. and Mrs. Billington. It was collected by them at Tshumbiri in 1905.

This interesting species may be readily recognised from all other members of the genus, by the curious spotted or mottled appearance of the thorax, pleurae, and femora. To the unaided eye it looks very like a dark specimen of *Glossina palpalis*, R.D.; but a pocket lens immediately reveals the peculiar markings.

FURTHER NOTES ON THE DISTRIBUTION OF TSETSE FLIES

Trustworthy persons frequently stated that tsetse flies of small size (not *fusca* or *longipennis*) existed in various localities. They were, however, unable to further identify the fly, and did not procure specimens. We have therefore compiled the following supplementary list of places in which tsetse flies occur. The names occurring in it have been considered in the preparation of the map showing the distribution of the *Glossinae*.

Matadi; Loeka; Redjaf, Lado Enclave (reported, Dr. Bignami); Makanga, Maniema (reported, April, 1905, Chef de Poste); Bene Kamba, on the Lomami River (recognised by natives); "All along the road from Kasongo to Baraka" (reported 1905, Bishop Roelens), confirmatory reports have been received from the following places along this route: Piani Lusangi, Kabambare, Kalembelembe, Yamba-yamba; Kibonga, Lake Tanganyika (reported, Chef de Poste); Lake Kivu (reported, Commandant Crone); Upper Luama River (reported, Bishop Roelens); "Places where tsetses do not exist in the Katanga district are so few as to be negligible"† (reported Feb. 4th, 1905, Monsieur Baillon, Chef de Secteur). Places in Katanga at which

* The abdomen is curved considerably, but this has not been allowed for in the measurement.

† This statement is in every way supported by numerous reports received by us, a few of them are cited, and by a recently published pamphlet entitled "La Mouche Tsetse et la Colonisation au Katanga, par le Lieutenant Brohez 1905, published by Vanderauwera et Cie, 59 rue de la Montagne, Brussels."

tsetses are definitely said to exist are: Ankoro; along the Lubili River; on Lake Tanganyika at Mpala and Vua;* Lusaka; Kiambi; Mpweto; Lukonzolwa; Kilwa; Lukafu; Kasenga; Kambove; Tenke; Shiniama; Kalonga and along the Lualaba, Lufira and Luapula Rivers; Upper Kasai and Luebo Rivers (reported, Mr. Verner); Angola (reported, Bastian, Interpreter at Lusambo); Popokabaka (reported, Dr. Hollebeke).

Map II has been prepared to show the distribution of *Glossinae* in the Congo Free State. For the sake of uniformity we have used the same signs as Mr. Austen to indicate the various species of *Glossinae*. As will be noted, our observations confirm his map in many instances and amplify it in others. In it has been incorporated the information contained in the article on tsetse flies by Austen, published in Volume VI of the Reports of the Sleeping Sickness Commission of the Royal Society and in a communication, on the same subject, made by Laveran to the Academie des Sciences.†

Remarks.- An important deduction to be made from the above notes on the *Glossinae* is that *Glossina palpalis*, at least, cannot be said to be absent from a given spot until several careful searches have been made at considerable intervals of time.

A careful study of the bionomics of tsetse flies is still urgently needed. Comparatively little is known at present concerning their habits; especially lacking is accurate information on the variation of their numbers in various localities ("fly belts") and on the causes governing their appearance or disappearance. On only one occasion did any hint of a natural enemy of these flies reach us.

The Rev. Mr. Grenfell had slightly wounded a bird (a darter?—native name, *ullinga*). It was taken living into the canoe in which he was travelling, and it was seen to eagerly catch the tsetse flies flying about the native paddlers.

GENUS STOMOXYS

Stomoxys calcitrans, Linn.

(Pl. iii, fig. 4; Pl. v, figs. 1-8)

Localities:- Lulango; Lusambo; in the Gambia at Bathurst and McCarthy Island.

* Dr. Noble has recently identified the tsetse flies "plentiful at Vua" as *G. palpalis*.

† "Comptes rendus des séances de l'Académie des Sciences, t. cxli, p. 929, 4 décembre, 1905."

Circumstances of capture:—This fly was caught in the open and in the houses of Europeans. It feeds on all mammals, but seems to be especially fond of feeding at the tips of dogs' ears. Both on the Gambia and on the Congo dogs were often seen with their ears raw and bleeding from the attacks of this pest.

*It is, I believe, over 60 years since Bouché discovered the larvae of *Stomoxys calcitrans* in warm stable-manure. Since that time no additional information on the habits of this insect has apparently been given†; and the characteristics of the earlier stages and also the metamorphoses have remained practically unknown. It was resolved therefore to:

1. Trace out the life-history and metamorphoses by rearing the insect from the egg; and
2. To find the natural habitat for the eggs, larvae and pupae.

The first was accomplished with comparatively little trouble; but the finding of the natural habitat for the earlier stages of this insect proved no light task, and involved a considerable amount of time and diligent searching. In the end, however, I succeeded in solving these problems; and it is hoped that these notes will not only prove of interest, but also of some practical value to those students who are engaged in the study of insects in connection with the transmission of disease by blood-sucking flies.

During the month of August an unremitting search was made for the larvae and pupae of this insect in the faeces of nearly all the domesticated animals, both in the farm-yards and the fields, but without success; manure heaps both old and recent were also searched, but none were discoverable. A number of females were

*Newstead. Reprinted from the Journ. Econ. Biol., vol. i, No. 4, pp. 157-166, pl. xii, 1906.

† Howard, whose excellent paper on "A contribution to the study of the insect fauna of Human excrement" (Proc. Washington Acad. of Science, Vol. II, p. 578) I had overlooked, says:—"Our first experience with the breeding habits of this fly was in 1889, when studying the horn fly of cattle. August 20, 1889, four specimens of *Stomoxys calcitrans* were reared from horse manure collected at Washington, and on the 19th of the same month large numbers of adults were observed by Mr. Marlatt, attracted to freshly-dropped horse manure. January 20 and May 14, 1890, other specimens were reared from horse manure, the last lot of manure being collected on November 27, 1889, so that the insect hibernates in either the larval or pupal condition (probably the latter) in or just under old manure.

"This species has not been bred from human excrement, but has been observed in out-of-door privies at Snickers Gap, Virginia; Alexandria, Virginia; and at Charlestown, West Virginia."

then captured and placed in a large cage well supplied with light and air, and fresh faeces of the horse, sheep, and rabbit. A small percentage of the insects laid their eggs under these conditions; but the eggs were invariably removed to receptacles which were more convenient for the study of the metamorphosis. It was found that two important conditions were necessary for the development of the larvae, viz., an almost complete absence of light and *an abundance of moisture*. Such conditions as these could only obtain in a state of nature in large faeces in shady or damp situations, or in heaps of manure.

Towards the end of September the earlier stages of this insect were found under natural conditions, and the facts relating to them are given under the heading of additional field notes.

HABITAT.—Farm-yards and stables are evidently the favourite haunts of this fly; it occurs also in the fields, parks, and open woods, especially where cattle are grazing, but is much less numerous in such places. It is evidently also by no means uncommon in some of our large towns, and numbers were seen at rest on the shop-fronts in the main streets of both Liverpool and Chester. It is fond of resting on surfaces fully exposed to the sun, such as doors, gates, and rails, and to a less extent also on stone and brick walls. Painted surfaces are also attractive to it, and the greatest number seen congregated together were disporting themselves on the sunny side of a red-painted iron tank at the old Chateau de Goumont, Waterloo, Belgium. They are very active; but their flight is quite inaudible at a short distance, the noise produced being very feeble. When disturbed they frequently return to the same spot, but more especially so in favourite resting-places. At night they retire to some sheltered spot, and numbers may be found at rest on the beams and rafters in open sheds in farm-yards, where they remain, almost inert, till the morning sun tempts them out again. They will also occasionally enter stables in the day time, and they were seen to enter such places through a narrow opening or a crack in the door.

They frequently clean their wings when in captivity, and this is accomplished with great precision, the hind legs being used for this purpose. The under surface of the wings are first combed, then the upper, the legs are then rubbed together, and the process is again repeated in exactly the same order.

POSITION WHEN AT REST.—The front part of the body is often slightly raised, but not invariably so; and the wings, which invariably touch at their bases, are widely divergent and carried in a horizontal position, lying practically in the same plane as the abdomen.

RATIO OF SEXES.—During the heat of the day the males preponderated;* but towards evening the sexes occurred in about equal numbers; the captured females were, however, nearly all freshly emerged ones, and a large proportion of the eggs which they laid in captivity proved infertile.

FOOD OF THE ADULT.—During a period of 14 days a careful watch was kept on both cattle and horses in various farm-yards where the flies were common, but no flies were seen either to alight upon the animals or to suck blood from them; at the same time several examples of both males and females were captured which were fully engorged with blood. Moreover, a freshly emerged male readily sucked blood from the writer's own hand. There is, however, no lack of authentic evidence as to the blood-sucking habits of this fly which need not be repeated in this communication.

Two specimens were seen to settle on fresh cow dung, and apparently feed upon the moisture on it, passing the extended proboscis rapidly over the surface; such habits were apparently exceptional or rarely seen in a state of nature; but in captivity they readily fed on the fresh faeces of the horse and sheep, more especially so on the latter. A female was also seen to drive its proboscis into the thorax of a dead companion and apparently suck up the juices of its body. Three specimens fed upon some sugar and water, and some also sucked up the moisture from a decayed and fetid potato.

MOVEMENTS OF THE PROBOSCIS.—The base of the proboscis is frequently depressed so that the palpi become fully exposed, but the tip of the labium remains practically in the same position, or is very slightly elevated. The elbowed joint in front of the palpi is very flexible, and can be instantly inflated so that the entire proboscis can be completely straightened, and either extended horizontally or depressed vertically. The labella can also be:—

1. Straightened, taking the same plane as the labium proper. This is the normal resting position.

* The ratio was about 3 males to 1 female.

2. Curved upwards and outwards, with a quick alternate movement to either the right or left, or repeatedly and rapidly curved to one side only. As the labella curve upwards the *anterior* portion of the labium also gives a distinct lateral twist, so that the dorsal groove is presented laterally.
3. The teeth of the labella can be curved outwards and ventralwards, giving them a bilobed appearance; the teeth also apparently move rapidly backwards and forwards, and a clear fluid was seen to pass down the tube when the insect was slightly pinched between the fingers.

In sucking blood from the writer's hand the insect sat high upon its legs, but the anterior pair were much elbowed, and all the joints of the tarsi generally rested upon the skin of the host. The whole of the proboscis was straightened and held vertically, and the anterior third was driven into the flesh.* During the process, which lasted altogether for a period of 15 minutes, the proboscis was constantly, but somewhat slowly, moved up and down, and also with an occasional semi-rotary movement, reminding one somewhat of the action of a quarryman's hand drill. This action was continued until the fly had pumped its body full of blood. The initial pain was trifling compared with that of a mosquito; but there were two subsequent pricks which were quite as irritating as the first. A small drop of blood was left over the puncture, and when this was washed away a small roseala was revealed; but there was no subsequent irritation or soreness of any kind. A clear fluid was passed from the anus four times during the process, and on several occasions subsequently, and judging from the size of the abdomen the food was rapidly assimilated. This fly died twelve hours after feeding.

DURATION OF LIFE.—In captivity they lived for several days; but they were supplied with abundance of fresh air and some moist faeces. The females died either immediately or shortly after laying their eggs.

EGG-LAYING.—When the female is about to lay its eggs the ovipositor becomes fully extended, and nearly equals the length of the abdomen proper. The eggs are passed rapidly down the ovipositor at intervals of a few seconds,† and were usually laid in an irregular

*In thick-skinned animals the proboscis would in all probability be driven in still further.

† 5-30 seconds.

heap. In some instances the female was seen to separate the eggs by carefully passing her proboscis between them, and then drag them away or scatter them with her legs. In cases where the eggs were laid during *extremis* the female generally died on the spot, and made no attempt to scatter them. Counts were made of seven batches, the maximum being 71, the minimum 48; the actual counts were 48, 48, 54, 57, 59, 62, 71. The incubation period, at an average temperature of 72° F. in the day and 65° F. at night, was two to three days.

TIME OF APPEARANCE.—They are especially abundant during August and September; but gradually diminish in numbers during the early part of October;* and few examples are seen after a spell of cold wet weather.

THE EGG.—Coriaceous; white at first, but changing to creamy white. Those which were laid on faeces, fully exposed to the sun, had the exposed portions tinged with pinkish-brown, but this colour eventually disappeared. Form very elongate, shaped somewhat like a banana, being curved on one side and almost straight on the other; the straight side with a broad deep groove which widens at the anterior end giving it a spatuloid form. Surface with faint polygonal reticulations. The larva effects its escape by splitting the broad end of the groove, leaving it slightly raised (see Pl. v, fig. 4), and apparently intact on the opposite side.

Length, 1 mm.

THE LARVA.—Colour, creamy-white to pale ochreous, translucent, shining and almost glass-like; sub-cutaneous mouth parts black; the convoluted alimentary tract when filled with food gives the posterior half a blackish or greenish-black colour; tracheal tubes forming two-submedian white lines and delicate lateral branches. Posterior stigmata black; thoracic stigmata ochreous. Form long, tapering to a point in front, widely rounded posteriorly. Segmentation not very pronounced. Epidermis without hairs. Head with two large divergent mammiform processes, at the extremity of which are the minute retractile antennae, apparently of four sub-equal segments. The mouth armature consists of five distinct parts: a strongly falcate hook in front, which articulates with a broad, thick and somewhat rectangular hypostomal sclerite; immediately below the great hook is

* A single specimen was seen in Liverpool on a bright sunny day during the first week in November.

a small dentate sclerite; the hypostomal sclerite articulates with two large bifurcated cephalo-pharyngeal sclerites, and in front of the upper arms of these pates is a small perforated sclerite. In a freshly prepared specimen both the retractor and extensor muscles to these sclerites can be distinctly traced. Ventral surface of the last seven segments furnished with raised bands of tactile tubercles. Posterior stigmata two in number, circular; thoracic stigmata placed sub-laterally on the third segment, each consisting of apparently five circular orifices, these are connected posteriorly with a large bilateral air sac which extends along the fourth segment.

Length of adult, 11 mm.

Young larvae are much more transparent and glass-like, and the large anterior mouth-hook is not developed, a blunter process taking its place.

HABITS OF THE LARVAE.—They move rapidly along a smooth surface, pulling themselves along chiefly by means of the large mouth-hook; and proceed practically in a straight line, moving the head rapidly but irregularly from side to side or up and down. There is, however, no regular alternate movement of the head during progression as in some muscid larvae. Their progress through the burrows in their food is much more rapid than on a smooth surface, and when disturbed they disappear with extraordinary rapidity. The larval stage lasted, under favourable conditions, from 14 to 21 days; *but the absence of excessive moisture* and the admission of a little light materially retarded their development, which then extended over a period of from 31 to 78 days.* The larvae exposed to such conditions produced much smaller pupae and correspondingly small imagines.

METHOD OF PUPATION.—This is completed in about two hours. At first the larva rapidly shortens itself, chiefly by contracting the anterior segments, and becomes barrel-shaped. At this period it is of a creamy-white colour, and the mouth parts of the larva are still visible through the soft integument. The colour rapidly changes to bright ochreous, and in the space of two hours or even less, the integument hardens and the puparium assumes its normal colour. In cases where soil was placed below the faeces the larvae generally burrowed into the former to a depth of about half an inch, but a few also pupated

* A few specimens still remain in the breeding cage, and may possibly pass the winter in this stage.

in the dryer portions of the dung. Where no soil was provided the larvae generally pupated at the bottom of the breeding cage.

THE PUPARIUM OR PUPA. Colour bright terra-cotta red, changing to dark chestnut-brown a few days before the emergence of the fly. Form barrel-shaped, slightly narrowed in front, broadly rounded behind. Eleven segments only are visible, the anterior one bearing the minute bilateral thoracic stigmata of the larva; posterior segment with two large disc-like stigmata; all the segments with fine transverse striae, the striae, under the microscope, producing a slight irridescence; dorsally the articulations have a double series of minute papillae, one series being more minute than the other; the posterior segment has also a median longitudinal series which terminates between the stigmata; ventrally the papillae are replaced by a regular series of fine ridges, forming a distinct and relatively broad band; the last segment also bears a somewhat lunular or angular-shaped patch of more or less rounded papillae. This stage lasted from 9 to 13 days.

Length, 5 to 5·50 mm.

The larvae fed on comparatively dry faeces produced much smaller pupae—3·50 to 4 mm.

DEVELOPMENT OF THE NYMPH.- A few days before the emergence of the insect the cuticle of the puparium darkens and eventually splits anteriorly along the lateral and median lines and also transversely along the fourth segment; the section falls away, and the fly escapes. Prior to this the nymph undergoes its final ecdysis, pushing its effete skin off backwards into the posterior end of the puparium. On its emergence it appears as a small dark-grey fly, with thick rudimentary wings of a dull leaden colour and a deep notch in the mid costa, below which are strong convoluted folds. The head is much larger and wider than the thorax, and the abdomen is attenuated. Its subsequent development may be conveniently divided into the following stages:—

1. An extremely active period, which lasts for approximately half an hour. During this period the insect devotes nearly the whole of its attention to the escape from its environment. If placed in a glass tube with a barrier of loose cotton wool in it, the fly immediately endeavours to effect a passage through it, and this it accomplishes with marvellous rapidity,

making headway by constantly inflating the frontal sac, at the same time pushing itself forward with the legs. When liberated, a great deal of time is devoted to combing out the hairs on the arista of the antennae, this being accomplished in the following way: the head is turned either to the right or left, as desired, and the frontal sac is then inflated on that side farthest from the thorax; this process lifts the antennae into a prominent position, and the long hairs of the arista are then rapidly and carefully combed out with the under surface of the anterior tibiae. The frontal sac also receives a share of attention, and so also does the abdomen, and occasionally the rudimentary wings. This stage is remarkable, in that nature so provides that, under normal conditions, the insect may successfully escape from its larval habitat before the wings develop, and so impede its progress or render its escape impossible.

2. In this stage the frontal sac is usually contracted, and the head presents a more normal condition; the fly also becomes quiescent, and remains as a rule in a fixed attitude, with the legs well displayed, and the head extended forwards, so that the narrow neck is stretched to its fullest extent. Air is then pumped into the body by repeated and alternate contractions and extensions of the abdomen. The body increases in size, and the integument becomes *extremely pallid in colour*. At this stage the wings are apparently filled with air, which passes rapidly along the costal region, then across to the hind margin, and finally the tip unfolds, sometimes aided by the use of the hind legs. The first portion of this stage sometimes occupies over twenty minutes; but the wings develop as a rule in about three minutes.

3. The fly still remains more or less quiescent, but gives some attention to cleaning itself, and when the integument and the wings are sufficiently hardened the proboscis is raised from the ventral to its normal horizontal position; when this is accomplished the insect takes flight.

Defaecation takes place shortly after the imago is perfected; the faeces being milk-like both in substance and colour.

Summary of Life Cycle.

Larvae fed on moist sheeps' dung.

Eggs procured from captive females.

Average day temperature	72° F.
Average night temperature	65° F.
Month	August.
Ova—Incubation period	2 3 days.
Larval stage	14 21 „
Pupal stage	9 13 „
Complete cycle	25—37 „

Summary of Life Cycle.

Food allowed to partly dry and some light admitted.

Temperatures as above, during the month of August.

Ova—Incubation period	2 3 days.
Larval stage	31 78 „
Pupal stage approximately as above,	
Complete cycle	42 78 „

Some larvae of this brood may hibernate through the winter.

Additional Field Notes.

While this paper was going through the press I continued my search for the larvae at various places within a few miles radius of Chester, and on 21st September my efforts were at last rewarded by finding the insect in all stages of its metamorphosis. The habitat was on the outside of a cucumber bed, at Rossett, Flintshire. Here both males and females were disporting themselves on the cucumber frame and also on some matting used for covering the glass. My attention was first given to an examination of the stable manure forming the hot-bed, but this was quite unproductive, as I ventured to think that it would be, owing to the rather dry nature of the outer walls. But lying alongside the cucumber bed was a heap of grass-mowings which had accumulated during the season. The uppermost layer had only recently been deposited, and this was quite hot (90° to 98° F.), below this the grass was quite rotten and sodden with moisture, but registered a temperature of 70° F. It was in this layer that I found the

first lot of larvae. They were chiefly full-fed, and some of them pupated during the next two or three days. Continuing my search further I also found numbers of larvae and pupae, in the still older and quite cold deposits, some of them were only a few inches below the surface, others were deeper down; they sometimes occurred singly, in other cases several were found together, some were mature, others only partly developed. On disturbing the newer deposits they naturally gave off the strong smelling fumes characteristic of heated grass, and this produced a result which I had neither hoped for nor anticipated. A female Stomoxys was seen to alight on the hot and freshly disturbed grass and to quickly disappear among the interstices, there she remained for a minute or so and then flew away. The grass was carefully examined, and amongst it, at a depth of nearly three inches, were found a number of her eggs. A regular succession of females then followed, and very soon three of them were engaged in laying their eggs in a small area which could have been covered with a crown piece; the first-comer being not in the least disturbed by a companion running completely over her body. In all cases the abdomen was depressed, and pushed into the material as far as possible, and in two instances the wings were partly extended, in order, apparently, to secure a firmer support. One female remained in the same spot for five minutes, and then changed her position to another a few inches away. A second female was occupied for twelve minutes in laying her eggs, but she did not change her position during the time. All three females flew away immediately afterwards, and did not seem in any way weakened by the process. For how long they survive in a state of nature it is impossible to say, but some females that were caught immediately after egg-laying died on the fourth or fifth succeeding days. Some females which were caught in glass tubes as they alighted on the grass laid their eggs immediately afterwards, and these also survived until the fourth and fifth days.

Reference has already been made to the almost noiseless flight of these flies when disporting themselves over and about their favourite haunts during the heat of the day. But I found that when the females were negotiating the habitat previous to laying their eggs the noise was distinctly audible, and resembled the characteristic hum produced by other muscids.

The day on which the foregoing observations were made was a delightfully bright and sunny one, and the hour from 3 to 3·45 in the afternoon; later, when the sun had lost its power, the flies disappeared, in their usual way, to find some sheltered spot in which to pass the night.

The eggs which were procured on this occasion were kept at a temperature varying between 64° and 67° F.; under these conditions the larvae did not begin to hatch until the *eighth day*, thus the incubation period was greatly prolonged; had they been left in the warm grass, where the temperature near the surface was 70° F., they would in all probability have hatched, as they did in the summer months, on the second or third day.

Whether the larvae of these autumn broods will pupate before the winter is at present impossible to say, but judging from the high temperature of the habitat, which will certainly be maintained for a fortnight at least, it is reasonable to assume that they will do so, and we shall probably find that the winter is passed chiefly in the pupal stage.* Any material disturbance of the habitat in question would result in complete loss of the artificial heat and a sudden check to the development of the larvae. In such a case the larvae would probably hibernate through the winter and pupate in the following spring or early summer. Fortunately this is not likely to obtain in this instance, as the owner has very kindly given me undisturbed possession of the whole of the material, so that it will, it is hoped, be possible to continue the observations through the winter and spring, though little of economic importance can now be added to the habits of the insect in this country.

Stomoxys sitiens, Rond.

Localities:—Nouvelle Anvers; Lulongo; Nyangwe; Kasongo.

Circumstances of capture:—Specimens were caught about cattle and in a European house. It feeds vigorously.

Stomoxys sitiens ?

A large series of specimens were caught on cattle at Zambie. They were preserved in alcohol and cannot therefore be definitely fixed.

* On Jan. 13th, both larvae and pupae were found in this habitat; the former were quite dormant, but became active in a warm temperature. Both stages occurred in the decayed grass about 6 inches below the surface.

Stomoxys omega, n. sp. (Newstead)

(Pl. iii, figs. 2, 3)

Wings dusky, strongly irridescent in a bright light. Thorax blackish, shining, anterior half in front of transverse suture pale greyish-blue, with a distinct ⋂-shaped black design in the male; in the female the submedian black lines blend with the black colour in front of the suture so that the up-curved terminals of the ⋂-shaped markings become indistinct. Abdomen, smoky-brown.

MALE.—*Head:* Eyes almost meeting, the narrow space between them, above, velvety black; frontal margins and clypeus silvery-white. Basal segment of antenna blackish; second segment brown; apical segment silvery-grey; arista brown; posterior surface of head velvety-black; lower angles silvery-white with black hair. *Proboscis* black, shining; labella with many, very long, fine outstanding hairs. *Thorax* in front of the median transverse suture grey-blue, with a large ⋂-shaped black design, consisting of two distinct broad submedian longitudinal stripes which curve upwards before reaching the suture and form two conspicuous rounded dilations; thorax *below the transverse suture* black, shining, with a rather broad band of dusky grey-blue in front of the scutellum, which is continuous to the base of the wings; scutellum smoky-brown, shining; with one central and two lateral black bristles; the thorax is also clothed with fine blackish hairs, and there are three bilateral black bristles at the sides in front of the wings; pleurae in front delicate grey-blue, a small area at base of wing pale brown. *Abdomen* hairy; smoky-brown, the segments gradually darkening towards the apical margins, especially so at the sides; in perfect specimens there is a broad basal band on the second, third and fourth segments of faintly greenish-grey dust, each band is interrupted in the middle; the last segment almost entirely clothed with a paler grey dust; venter pale dusky-brown. *Legs* relatively long, smoky-brown, blackish in some lights; knees pale brown.

Length, exclusive of proboscis, 6 mm.; length of wing 7 mm.

FEMALE.—*Head* with the space between the eyes relatively narrow; vertex, blackish, shining; margins and clypeus pale bluish-grey, bright but scarcely silvery; posterior surface of the head similar to that in the male, but the sides below have a broader margin of blue-grey. Apical segment of *antenna* dusky-grey. *Thorax* in front

of the median transverse suture grey-blue, with two short, well-defined submedian black lines in front, these gradually merge into large and somewhat ill-defined black areas, leaving the blue-grey ground visible only at the sides and between the black longitudinal bands; thorax behind the suture as in the ♂; scutellum black, paler in the centre, shining; pleurae and legs as in the ♂. *Abdomen* similar to that in the male but *without the grey, dust-like margins.*

Length, exclusive of proboscis, 6 mm.; length of wing 6·50 mm.

Easily distinguished by the beautiful pale blue of the anterior half of the thorax, with, in the ♂, the distinct black ∩-shaped pattern.

The relatively narrow space between the eyes in both sexes is also noteworthy, these characters being much more pronounced than in the species of the *calcitrans* type.

Localities:—Ukungwa; Sendwe.

Circumstances of capture:—This fly was seen only near water. Some were caught in canoes, others on a buffalo shot in a marsh, on which they were feeding. When the buffalo was first seen it was lying half-covered in water, no doubt to avoid the *Stomoxys*, which were present in almost incredible numbers.

CUTANEOUS MYIASIS

? Ochromyia anthropophaga (E. Blanch.), in man.

We are greatly indebted to Dr. Grenade of Leopoldville for three specimens of a larva or "bot" closely resembling that of an oestrid fly, taken from a coloured patient in hospital at Leopoldville. Two of them occurred in the scalp, the other in the neck.

These larvae agree so well with the description given of *Ochromyia anthropophaga*, E. Blanch, that they possibly belong to this species or a closely related one.

Larva of a Muscid Fly in a Rat

The following description of a larva or maggot taken from the tail of a tame white rat at Kasongo, February 16th, 1905, may eventually lead to the identification of the insect; so far we have been unable to fix it.

Description.—Length 11 mm. Cylindrical, but slightly flattened; rather suddenly tapering in front, rounded behind but *not truncate.*

Segmentation very pronounced and regular above but convoluted ventrally; all the segments, with the exception of the antepenultimate which carries the anterior stigmata, almost covered with very short and rather blunt spines, the majority of which have dark brown or piceous tips; these spines are arranged in short straight transverse lines, each group consisting usually of from three to five spines, in some cases there are but two and in others more than five. The great mouth hooks are black; and there is a lateral palmate group of external teeth as in the larva of *Auchmeromyia luteola* (q.v.). Posterior stigmata on the last segment, subdorsally placed, and very close together.

Further details cannot be given, as it has been thought well to preserve the specimen intact.

Larvae of a Muscid Fly in a Mule

The mule illustrated in fig. 16 was brought to us at Kasongo. In the centre of a large and very conspicuous oedematous swelling on its abdomen was a deep-seated ulcer, some three inches in diameter. On inspection, the whole ulcer, below the level of the epidermis, was found to be so closely set with the larvae, described below, that it was impossible to see any part of its base.

The mouth parts of the larva were invariably directed inwards and the bodies of the parasites were so deeply buried in the tissues of the host that their posterior extremities were alone visible. It was found that there were two layers, one above the other, of larvae, packed side by side closely together. It seems probable that they were able to burrow in the tissues of their host, since a few larvae were found in the sloughing tissues at a depth of some five centimetres from the apparent surface of the ulcer. None of the larvae penetrated into the abdominal muscles. About 160 to 180 larvae were taken from this lesion, which healed quickly after a thorough curetting and dressing with iodoform. A description of the larva is herewith appended:—

Length 11 mm.; greatest width 2·25 mm.

Colour, in formol, dull pink, with the anterior segments paler; some examples were almost white. Form spindle-shaped, tapering from the mid region almost to a point in front, and slightly so posteriorly. Great mouth hooks prominent, unidentate, and black. Segmentation very pronounced, each with a strongly marked ridge forming

a complete ring round the body; all the ridges furnished with short brownish-coloured spines, which to the naked eye appear as distinct equidistant rings; anal segment somewhat truncate, but deeply wrinkled, and furnished dorsally with four very short papillae and ventrally with two much longer ones. Anal stigmata, brown or dark castaneous, almost touching; irregularly ovate, with three transverse slits.

Attempts to raise the fly from these larvae were unsuccessful.

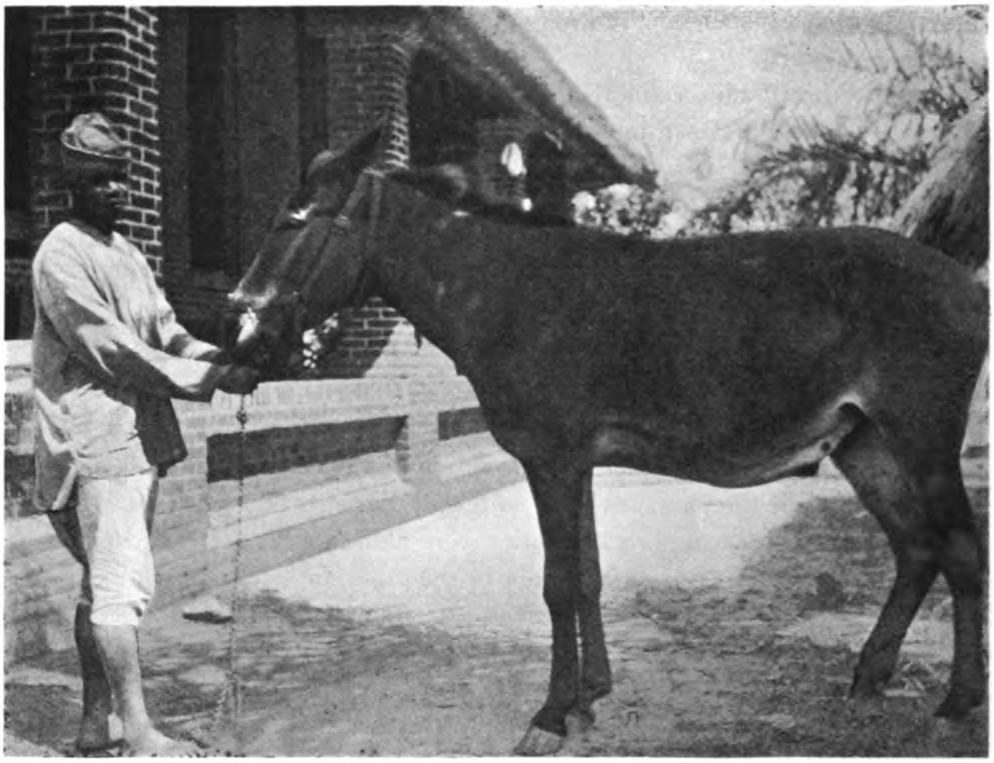


FIG. 16.—*Cutaneous myiasis.* Mule showing oedematous swelling on abdomen, containing larvae of a Muscid Fly.

FAMILY HIPPOBOSCIDÆ

Hippobocsa equina, Linn.

(Figs. 17, 18)

A few examples of this species were found on cattle, shipped at Las Palmas, Canary Islands, while on board ship on their way to the

Congo Free State (September 15th, 1903). This is an important record, and furnishes a further proof of the ready means by which these parasites may be introduced into a new and uninfected region.

Flies of this genus were also caught on camels and cattle at St. Louis, Senegal, May, 1903; their freshly deposited larvae were marked with very delicate tracings. Unfortunately the specimens and drawings were lost while in the hands of the Authorities of the British Museum and cannot now be traced.

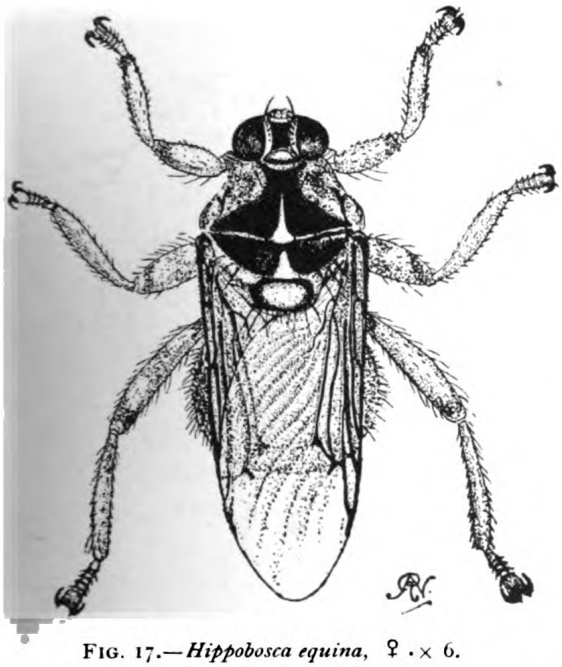


FIG. 17.—*Hippobosca equina*, ♀ . × 6.

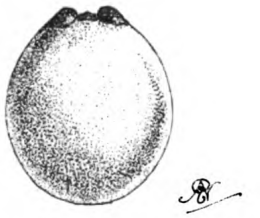


FIG. 18.—*Hippobosca equina* Puparium about twelve hours after extrusion. At this stage the anal tubercles are black; the rest of the integument terra-cotta red. × 6.

Lipoptena paradoxa*, n. sp. (Newstead).

(Figs. 19, 20)

FEMALE.—Specimens preserved in Canada balsam and alcohol are bright red-brown inclining to orange-brown at the sides of the abdomen; claws black; base of abdomen with a bilateral patch of darker chitin, the median area of the remaining segments also with darker markings, but these are both irregular and inconstant in the preserved examples. *Head* as wide as the anterior part of the

* *Lipoptera* of Siebold and Loew.

thorax; ocelli absent. Mouth parts rudimentary. Outer margin of eyes with a double series of spinose hairs. *Thorax* narrower in front than behind, with a submedian series of about nine long spinose hairs forming a curved line, and a short submarginal series of usually four similar ones terminating opposite the insertion of the mid legs; posterior margins with four long spinose hairs on either side of the scutellum; the last-named organ is also furnished with four similar hairs. *Abdomen* short ovate, almost sub-circular, with numerous spinose hairs arranged as shown

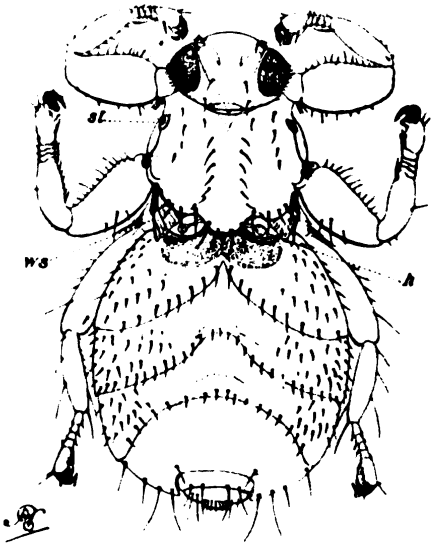


FIG. 19.—*Lipoptena paradoxa*, ♀ × 15. *st.* stigmen; *ws.* wing stumps; *h.* haltere.

in the figure. Venter with numerous short spinose hairs; median convex area with numerous minute equidistant tubercles bearing slender spinose hairs, the spaces between the tubercles finely but strongly rugose. *Legs* short, stout, sparsely clothed with hairs of varying lengths and varying degrees of thickness; the posterior pair not extending beyond the tip of the abdomen; tibial spine to anterior and mid legs stout; tibial spine to posterior legs long, slender; pulvillus broadly dilated from the middle outwards, finely spinose; feather-bristle strongly spinose; the upper surface with only one series of spines, the inner with two or three; ungues very faintly and irregularly toothed on the inner margin.

Length 4 mm.; width of abdomen 2 mm.

Habitat: Taken from an antelope at Kasongo, January 28th, 1905.

All four specimens are females. The same host also harboured a number of ticks. (q.v.)

The absence of ocelli in the female is rather remarkable. There is also an almost entire absence of *external mouth* parts, including the labial sheath; the only indication of these organs being a minute truncated cone, the exact nature of which could not be determined in the limited supply of material.

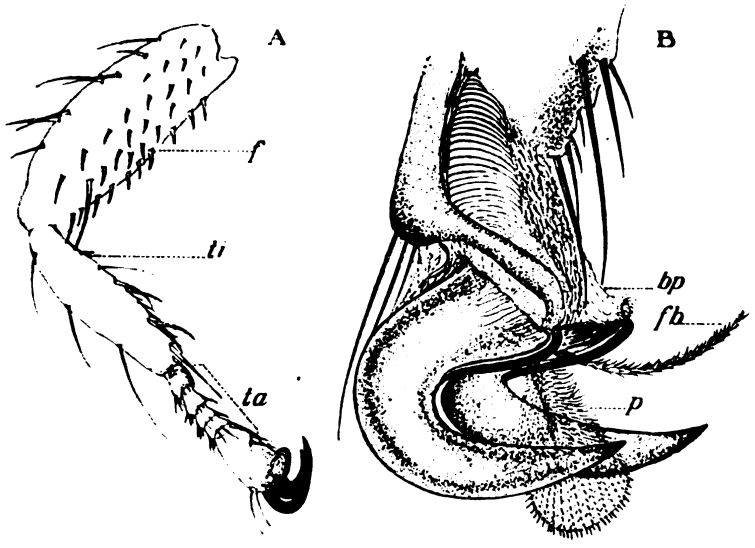


FIG. 20.—*Lipoptena paradoxa*, ♀. *A.* × 35; *f.* femur; *ti.* tibia; *ta.* tarsus. *B.* claws × 140; *bp.* basal process; *fb.* feather bristle; *p.* pulvillus.

FAMILY PULICIDÆ

Dermatophilus (*Sarcopsylla*) *penetrans*, Linn.

(Pl. vi, figs. 1, 2)

Larva long, cylindrical, of fourteen almost equal segments. Head slightly longer than the second segment, narrowest in front, but rather widely rounded. Antennae short, apparently of three segments, the basal and apical segments very short; apex with a central long slender spine and two to three minute ones. Surrounding the base of the antenna ventrally is a semicircular group of blunt spines, the two

nearest the base of the antenna much the largest. Mandibles large, slightly curved, unidentate. Cuticle below the buccal cavity with a curved row of short, stout, and backwardly curved spines; on either side of this series of spines are two divergent subcutaneous sclerites, the ventral one extending almost to the articulation of the head with the thorax; the upper sclerite about half the length of the ventral one; both are narrow and rod-like; the base of the ventral sclerite is also much dilated, and at the point where it meets the upper one it is unequally bifurcated and spine-like. Cuticle of all the segments presenting a strongly marked scale-like appearance; each segment is also provided with a single transverse series of long equidistant hairs, placed near the articulations; terminal segment bilobed, each lobe furnished with a single short stout spine and numerous hairs, these lobes are somewhat analogous to the anal claspers in certain Lepidopterous larvae.

Length 1·50 to 2 mm. These measurements are from apparently immature larvae, but seeing that the sexually mature insects are very small (1 mm.), it is highly probable that the larva does not attain much greater dimensions.

The number of segments given is inclusive of the head.

This pest is found practically everywhere in the Congo.

They were seen to be very plentiful at Nyangwe, and we were told that at Rutshuru and Beni they almost constitute a plague.

The larvae are extremely difficult to locate; and it was only after a prolonged search that we succeeded in finding three specimens. They occurred among the dust from the floor of a native hut which was literally swarming with chiggoes.

FAMILY PEDICULIDÆ

Pediculus capitis, De Geer.

This louse was only noted on cases of human trypanosomiasis at Leopoldville and Kasongo. Their distribution is most probably much wider.

Pediculus vestimenti, Leach.

These vermin were only noticed at Boma and Leopoldville; their distribution is very probably much wider.

ORDER HEMIPTERA

FAMILY CIMICIDÆ

Cimex lectularius (Common Bed-bug)

Localities:—Banana (Dr. Etienne); Tshumbiri (Rev. Billington); Nouvelle Anvers (Dr. Müller); Tshofa; Kabinda; Lusambo.

Circumstances of capture:—Specimens were taken from the crevices in the cane beds and grass-cloth pillows of natives.

ORDER HYMENOPTERA

Melipona, sp.

One or more species referable to this genus were seen in several places in the Congo Free State. They were particularly numerous at Leopoldville and Dibwe. They caused intense annoyance by the persistent manner in which they swarmed about one and crawled into one's mouth, eyes, nose and ears. When crushed they emitted a peculiar and characteristic odour.

At Leopoldville a colony of these stingless bees had taken up their quarters in a large lock and rendered it quite useless by the great accumulation of wax which they had formed. In this instance it was interesting to note a waxen tubular opening, about 4.c.m. in length and about the thickness of a pencil, projecting from the keyhole. A species of *Melipona* has been noticed to have very similar habits in the Soudan.*

INSECTS OF NON-ECONOMIC IMPORTANCE

In addition to the insects dealt with in the foregoing chapters of this Report, a large number of insects, representative of various orders, were also collected; but as they are of no economic importance it has been thought desirable to deal with these elsewhere. We would add, however, that among the more remarkable species is a new *Dejeania* which was discovered among the *Glossinae* in the fly cages at Kasongo. It had evidently been caught by one of the boys in mistake for an engorged tsetse, which it very closely resembles. We also obtained several specimens of a rather remarkable species of

* 2nd Report Wellcome Research Laboratory, Department of Education, Soudan Government, 1906, page 89.

Notocanthis of a uniformly blackish colour, with numerous white spots on the wings. These flies were only observed in one locality in the bush about a native village situated on high, dry, sandy soil, near Lisala (July, 1904). We possess no particulars concerning its habits. Whilst at Boma (Oct. 27, 1904) we also found a number of larvae or "rat-tailed maggots" apparently belonging to the genus *Eristalis*.

ORDER ACARINA

FAMILY HYDRACHNIDÆ

Ectoparasites of Mosquitoes

The larvae of four distinct species of *Acari* belonging to this family were observed on several species of mosquito. The site preferred by these ectoparasites was invariably the dorsal area of the abdomen, and generally, on the first few segments. Most of them, though not all, were fixed to the articulations of the segments, to which they seem firmly attached. The mosquitoes most subject to the attacks of these acarids were the following :—

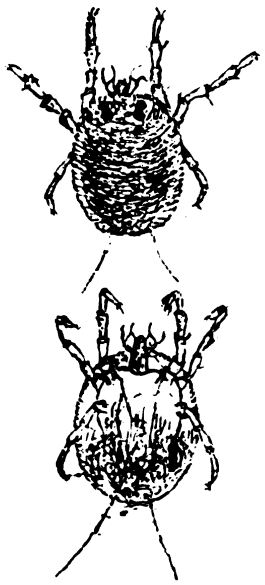


FIG 21.—*Ectoparasites of Mosquitoes.* × 20, about. From a sketch by the late Dr. J. E. Dutton.

Myzorhynchus paludis.—About 30 per cent. of these mosquitoes had the parasites attached to them. In one case as many as five were found clustered together on the articulation of a single segment. The species met with on this host (when dry) is of a bright ochreous colour with a dull orange scutum and rather large black eye-spots. The localities were Kumba, Kasonga and Lusambo.

Mansonia africana.—A large percentage of these insects also carried parasites, of which there were three well-marked species: one of a uniformly orange-red colour and another of a creamy-white or pale ochreous. In life some of these acarids were pink or yellow. Localities: Leopoldville, Tshumbiri, Kasongo and Lusambo.

FAMILY SARCOPTIDÆ

A case of Symbiotic mange occurred on a rabbit which was imported from Europe for experimental purposes. The condition was severe and the external ear was entirely filled with the parasites and the very profuse epithelial desquamation they had excited. On macerating the dead specimens in potash, one finds that they are a species of *Symbiotes* (now referred to the genus *Chorioptes*) and agree best with the descriptions given of *Symbiotes communis* var. *cuniculi*.

FAMILY DERMANYSSIDÆ

Pneumonyssus duttoni, Newstead and Todd

Memoir xviii, Liverpool Sch. Trop. Med. p. 4 (with plate), 1905.

Since the publication (l.c.) of the description of the adult mite Dr. J. W. B. Hanington has been successful in discovering the earlier stages of this acarid, and a description of them is appended below: -

Ovum.- Rather narrowly ovoid; faintly yellowish-white and, as seen by transmitted light, rather granular in appearance.

Length ·32 mm.

Hexapod larva.- Short ovate, narrowed in front, widely rounded behind. Capitulum prominent. *Palpi* of five segments furnished with short slender hairs with the exception of the apical one, which has several long stiff hairs; basal segment broadest, but scarcely longer than the apical one; second, third and fourth shortest and nearly equal in length; formula (1, 5) (234); tip of the apical segment reaching to the middle of the third segment of the anterior legs. *Legs* directed forwards; all the segments with whorls of hairs near the articulations, those on the basal segments are mostly short and slender, while those on the remaining segments are long and stiff; tarsi longer than the two preceding segments; all with very long hairs, and a few short slender spines; claws simple, placed on a long pedicel which is much narrower at the articulation; pulvillus scarcely wider than the dilated claws, but the end extends beyond them.

Length ·50 mm.; width ·30 mm.

FAMILY TROMBIDIDÆ

The larvae of an acarid belonging to the genus *Trombidium* were found in considerable numbers attached to the naked skin on the muzzles of horses at Kasongo. They appeared to the unaided eye as minute crimson specks, and were evidently the cause of an intense eruption. The skin was pink, rough, and dotted here and there with tiny spots of dried blood; still attached to the skin or on slightly raised hairs were numerous profuse crusts of desquamated epithelium and dried serum. Although a careful search was made the parasites were not found elsewhere on these animals nor on any other host in that locality.

The larval forms of these acarids are well known for the intense itching and soreness which they cause by burrowing under the skin of man and other animals. It is generally an unnatural position for the mites, and as a rule they soon die; though one species which is often found on the ears of cats in this country (Great Britain) lives for many days, causing great annoyance to the infected animals.

The adult mite is not parasitic but predaceous, living on small insects and their ova. The only adult form collected in the Congo (Leopoldville, Oct.) was *Trombidium grandidissimum*, a large spider-like creature completely covered with a brilliant crimson pubescence which is so dense as to appear quite velvety in texture.

FAMILY IXODIDÆ*

The fourteen species of ticks here catalogued include two that are new and undescribed.

The only member of the Argasidae which we met with was *Ornithodoros moubata*; this important species has already been described and figured in the Report on the nature of Human Tick fever.† We therefore only add here a few additional notes on its bionomics.

A few hundreds of these ticks were brought to Liverpool in September, 1905; they have since been kept in an incubator at a temperature of between 19° and 22° C., where they have reproduced freely, but under these conditions the eggs take two or three days

* This collection was very kindly identified by Prof. G. Neumann.

† Liverpool School of Tropical Medicine Memoir XVII (Plates and map).

longer to hatch-out than was noted in the Congo. Many of the original ticks are still alive; they have of course been well and frequently fed. One female raised in the laboratory from an egg has now been under observation for 25 months; during this period she has moulted six times. This seems to be rather less often than is usual, since ten out of fourteen ticks raised from the eggs moulted six to nine times during the year. Not infrequently ticks do not seem to be able to shed their old skin, and die while moulting. As a rule the ticks would not feed properly more often than every seven to ten days. Ticks remained alive on several occasions without food for four months, and in one instance for over six months. Adult ticks caught before April, 1905, or their progeny, which have never been fed upon an infected animal, are still (November 22, 1906) able to infect susceptible animals by their bites.

Amblyomma hebraeum (Koch), var. *splendidum*, Gieb.

Forty-three males and eight females were taken from a Buffalo (*Bos nanus*) which was shot in a marsh near Tshumbiri. In life the ground colour of this beautiful tick is of a soft green, with brownish ornamentation; there is little or none of the irridescence so conspicuous in specimens preserved in alcohol.

Amblyomma variegatum, Fab.

On cattle, Kasongo (Feb.). Associated with *Hyalomma aegyptium*, *Rhipicephalus sanguineus* and *Margaropus annulatus*.

Amblyomma variegatum ?, Fab.

A single male was caught on a bovine at Kasongo (Feb.).

Hyalomma aegyptium, L.

Three females were caught on cattle at Kasongo (Feb.). They were associated with the preceding species.

Haemaphysalis leachi, And.

This tick is the recognised carrier of Malignant Jaundice (*Piroplasma canis*) in dogs.

Thirteen males and two females were taken from a leopard at Tshumbiri by the Rev. Billington (June); and Dr. Etienne sent a single specimen (host not mentioned) from Banana (Oct.).

Margaropus annulatus, Say.

Common on cattle at Kasongo (Feb.), Zambie and Coquilhatville. The specimens collected were all females.

Margaropus annulatus, var. *calcaratus*, Bir.

In all the active stages, on cattle at Coquilhatville.

Rhipicephalus bursa, Can. and Fanz.

Collected by the Chef de Poste on cattle at Nya Lukemba to the North of Lake Tanganyika. A single female was also caught on a bovine at Kasongo.

This species is the carrier of Carceag or Malignant Jaundice in sheep.

Rhipicephalus capensis, Koch

Eight females and four males were taken on cattle at Nyangwe.

Rhipicephalus nitens, Neumann

Locality: One male and three females were taken from an antelope (*Tragelaphus scriptus*) shot at Kasongo (Feb.).

Rhipicephalus simus, Koch

This is said to be one of the carriers of *Piroplasma parvum*, Theiler.

Two males and one female taken at Kasongo (Feb.); and three males at Banana (Oct., Dr. Etienne). The name of the host was not obtained in either case.

Rhipicephalus sanguineus, Latr.

Locality: At Banana (Oct.), collected by Dr. Etienne, host not named; at Kasongo, common on cattle; occurs in association with preceding species and *R. longus*.

In addition to the above, two species of Ixodidae new to science were also discovered. These are described by Prof. Neumann in the succeeding paper.

ERRATA.

Plate iv. For Tabanide read Tabanidae.
Map ii. Delete "(see pages)."

EXPLANATION OF PLATE I.

MIMOMYIA MALFEYTI. (Page 29.)

Fig. 1. Head of female showing the character and distribution of the scales. × 25, about.

Fig. 2. Tibial spines of male. × 75.

Fig. 3. Genital armature of male (left half). × 75.

MIMOMYIA AFRICANA. (Page 28.)

Fig. 4. Head of female showing the character and distribution of the scales. × 25 about.

ANISOCHELEOMYIA QUADRIMACULATA. (Page 32.)

Fig. 5. Head of female showing the character and distribution of the scales. × 25, about.

Fig. 6. Proboscis of the female. Enlarged.

NEOMELANICONION PALPALE. (Page 31.)

Fig. 7. Head of male showing the character and distribution of the scales. × 25, about.

Fig. 8. A few segments of the antennae of the male. Enlarged.

Fig. 9. Male palpus. Enlarged.

ERETMAPODITES INORNATUS. (Page 12.)

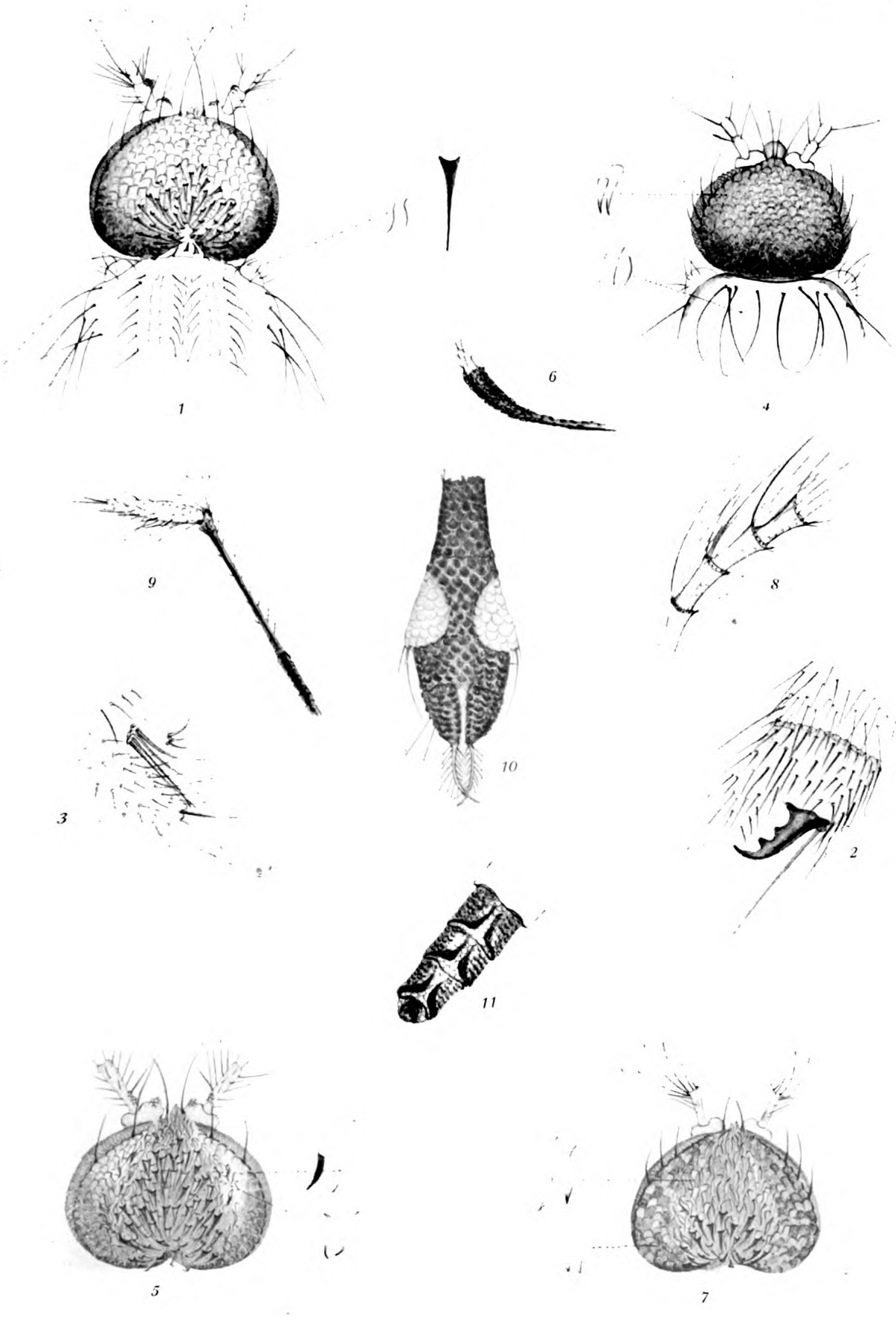
Fig. 10. Terminal segments of male abdomen. Enlarged.

CULEX PAR. (Page 25.)

Fig. 11. End of Abdomen of female (ventral). Enlarged.

All the figures are reproduced the same size as the original drawings from which they are taken.

PLATE I.



R. Newstead, ad nat. del.

P. P. Press, Imp.

EXPLANATION OF PLATE II.

BOYCIA MIMOMYIAFORMIS. (Page 34.)

Fig. 1. Head of female showing the character and distribution of the scales. × 25 about.

Fig. 2.—Head of male showing the characters and distribution of the scales. × 25, about.

Fig. 3.- Palpus of male. Enlarged.

STEGOMYIA ALBOMARGINATA. (Page 16.)

Fig. 4. Head of female showing the character and distribution of the scales. × 25, about.

STEGOMYIA LUTEOCEPHALA. (Page 15.)

Fig. 5. Head of female showing the character and distribution of the scales. × 25, about.

DUTTONIA TARSALIS. (Page 18.)

Fig. 6. Anterior tarsus of male. × 75.

Fig. 7.— Palpus of male. Enlarged.

Fig. 8.- Genital armature of male. × 25.

All the figures are reproduced the same size as the original drawings from which they are taken.

PLATE II.



R. Newstead, ad nat. del.

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EXPLANATION OF PLATE III.

TABANUS BILLINGTONI. (Page 46.)

Fig. 1. Female with wings displayed. × $2\frac{1}{2}$.

STOMOXYS OMEGA. (Page 87.)

Fig. 2. Female with wings displayed. × 4.
Fig. 3. Male with the wings omitted. × 4.

STOMOXYS CALCITRANS. (Page 75.)

Fig. 4. Female with wings omitted. × 4.
(In cutting away the background, the block-maker has inadvertently removed a portion from the left side of the abdomen. R.N.)

GLOSSINA MACULATA. (Page 73.)

Fig. 5. Female with the wings displayed. × 4.
Fig. 6. Female with the wings at rest (profile). × 4.

GLOSSINA PALPALIS. (Page 57.)

Fig. 7. Female in the act of parturition. × 4.
Fig. 8. Puparium before the escape of the imago. × 4.
Fig. 9. Puparium after the escape of the imago. × 4.

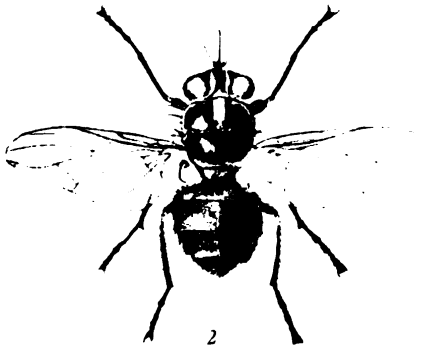
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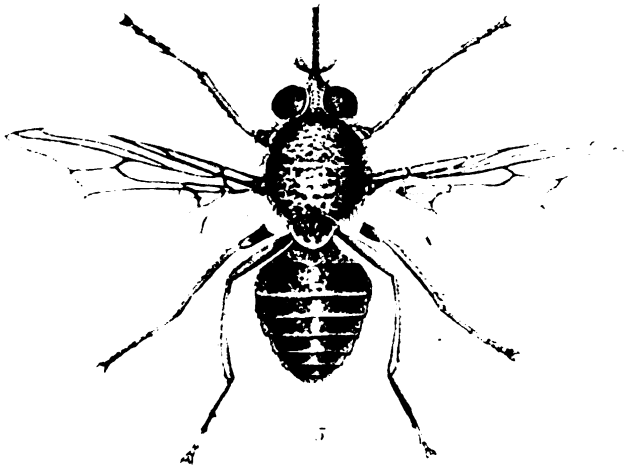
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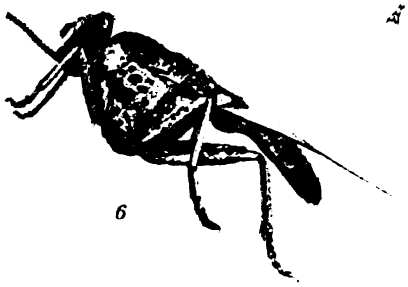
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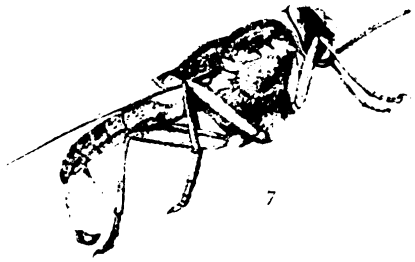
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R. Newstead, ad nat. del.

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EXPLANATION OF PLATE IV.

Fig. 1. CHRYSOPS DIMIDIATUS. (Page 43.)

Fig. 2. HAEMATOPOTA TRIMACULATA. (Page 42.)

Fig. 3. HAEMATOPOTA DUTTONI. (Page 41.)

Fig. 4. TABANUS DORSIVITTA? (Page 44.)

Fig. 5. TABANUS UNIMACULATUS. (Page 46.)

Fig. 6. TABANUS PAR. (Page 45.)

Fig. 7. TABANUS PLUTO. (Page 45.)

Fig. 8. TABANUS RUFIPES. (Page 45.)

Fig. 9. TABANUS CANUS. (Page 44.)

Fig. 10. TABANUS BILLINGTONI. (Page 46.)

Fig. 11. TABANUS BILLINGTONI. (Page 46.)

Fig. 12. TABANUS BILLINGTONI. (Wings displayed.) (Page 46.)

Fig. 13. TABANUS TARSALIS. (Page 45.)

Fig. 14. TABANUS FASCIATUS. (Page 44.)

Fig. 15. TABANUS GABONENSIS. (Page 45.)

All the figures are the actual size of the originals, photographed direct.



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15

AFRICAN TABANIDE.

R. S. Photo.

P. P. Press, Imp.

EXPLANATION OF PLATE V.

Illustrating Mr. Newstead's paper on "The Life-history of *Stomoxys calcitrans*, Linn." p. 75.

Fig. 1.—Eggs twice natural size.

Fig. 2.—View of the curved side of the egg. × 65.

Fig. 3.—Egg in semi-profile showing the deep spatulate groove. × 65.

Fig. 4.—Empty egg as seen in profile, with the semi-detached capsule at the anterior end. × 65.

Fig. 5. Dorsal view of larva showing the intestinal tract and course of the main tracheae. × 7.

Fig. 6. Three terminal segments of the larva in profile, with the internal mouth armature: *an*, antennae; *m*, muscles; *ps*, perforated sclerite; *md*, mandible or great hook; *hs*, hypostomal sclerite; *cs*, cephalo-pharyngeal sclerites; *vt*, ventral tooth. × 60.

Fig. 6A.—Chitinised mouth armature dissected out. Reference letters as in the preceding fig. × 60.

Fig. 7.—Three terminal segments of larva, dorsal: *cst*, compound thoracic stigmen; *as*, trachea forming internal air sac. Other reference letters as in fig. 6. × 60.

Fig. 8. Puparium or pupa. × 7.

EXPLANATION OF PLATE VI.

DERMATOPHILUS (SARCOPSYLLA) PENETRANS (Chiggoe Flea).
(Page 93.)

Fig. 1. Plantar surface of human foot with chiggers *in situ*. Actual size.

(*aa*) Group of eleven females.

(*bbbb*) Isolated females.

(*cc*) Pits or cavities left after the removal of the females.

(*d*) Section of epidermis with two females *in situ*. (Lateral view.)

Fig. 2.—

(*a*) Larva showing the squamose character of the epidermis. × 60.

(*b*) Outline of a younger larva. × 60.

(*c*) Antenna of larva with its accompanying group of blunt spines. × 250.

(*d*) Mandibles of larva. × 250.

(*e*) ? Cephalo-pharyngeal plate of larva. × 250.

(*f*) Buccal spines of larva. × 250.

(*g*) Empty cuticles of ova. × 60.



Fig. 1. Human Foot with "Chiggers" in situ.

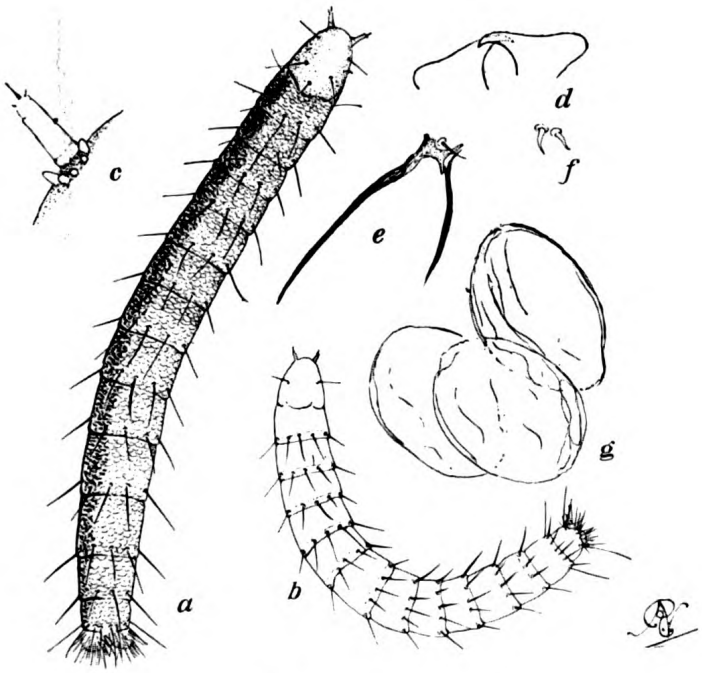


Fig. 2. Dermatophilus (Sarcopsylla) penetrans.
Details of Larva, etc.

R. Newstead, ad nat. del.

P. P. Press, Imp.

DESCRIPTION OF TWO NEW SPECIES OF AFRICAN TICKS

DESCRIPTION OF TWO NEW SPECIES OF AFRICAN TICKS

BY

G. NEUMANN

PROFESSEUR À L'ECOLE NATIONALE VÉTÉRINAIRE DE TOULOUSE

Rhipicephalus duttoni, n. sp. (Neumann)

MALE.—Body, narrow in front, broadest (1·85 mm.) a little posterior to the middle, length with rostrum 3·55 mm. *Scutum* slightly convex, chestnut-brown without spots, abdomen does not extend beyond its margins; cervical grooves are very broad, shallow, and form elongated depressions, they are not punctated and are continued posteriorly by a narrow superficial groove which extends beyond the middle point of the length; marginal grooves broad, shallow, slightly and finely punctated, commencing almost immediately behind the eyes and terminating in the groove which separates

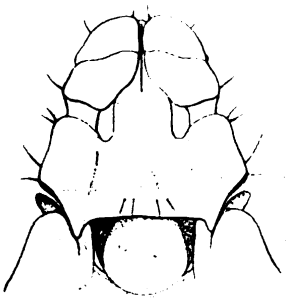


FIG. 22.—*Rhipicephalus duttoni*. ♂. Rostrum. × 45.

the two last from the following festoon; punctations irregular, coarsest in front, fine and superficial over the remainder of the surface; behind are three wide, shallow, unpunctated longitudinal grooves, the middle one being the longest; festoons longer than they are broad, slightly punctated, normal. *Eyes* flat, yellowish, large, marginal. *Ventral surface* reddish-brown, covered by rather long and abundant whitish hairs. *Anus* anterior to the middle of the

length of the adanal shields ; adanal shields have the shape of a scalene triangle and so form a long internal posterior spine (the internal edge is longest, it is rectilinear in its anterior half but is concave behind ; the external edge is slightly convex ; the posterior edge is concave and bordered by punctations) ; the outer shields are replaced by a prominent, non-chitinous fold ; no caudal prolongation but a chitinous thickening on the median festoon. Peritremes narrow, whitish, comma-shaped with the point bent back towards the dorsal surface. *Rostrum* 0·6 mm. long, dorsal base almost twice as broad (0·6 mm.) as long, lateral angles at about the middle of the length, posterior angles quite prominent. Hypostome very slightly spatulated, has six rows of teeth. Palps as broad as long, flattened dorsally ; second segment scarcely longer than the third and retracted into a blunt point dorsally at its

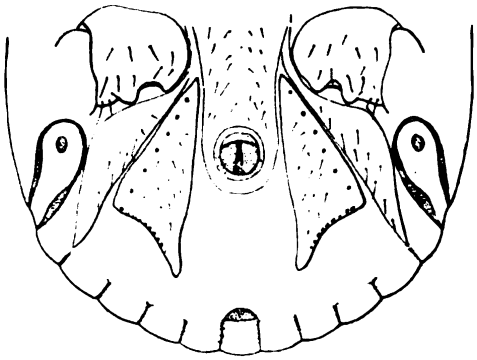


FIG. 23.—*Rhipicephalus duttoni*, ♂ . Ventral surface of posterior extremity. × 33.

posterior border. *Legs* relatively strong. Coxae covered with long white hairs ; anterior summit much elongated and conspicuous on the dorsal surface as an auricle, two very long spines ; on the posterior borders of the second and third an external spine, flat, as broad as long ; fourth divided at posterior border into two broad flat spines. Tarsi of medium size, have two terminal successive spurs ; caruncules large.

FEMALE unknown.

A single male was taken on a bovine at Zambie together with females of *Margaropus annulatus*.

This species is dedicated in honour of our late colleague Dr. J. Everett Dutton.

Rhipicephalus longus, n. sp. (Neumann)

MALE.—Body narrow in front, sides subrectilinear, length with rostrum 4·1 mm.; broadest (2·15 mm.) towards the posterior third. *Scutum* almost flat, dark chestnut-brown, without spots; along the posterior festoons the abdomen extends beyond its margin; cervical grooves, very short and deep; marginal grooves deep, narrow, each occupied by a row of punctations, commencing a little behind the eyes, ending at the posterior border of the penultimate festoon, and are continued forward, and especially inwards, by a row of coarse, scattered punctations; punctations irregular, abundant in the space enclosed between the marginal grooves and the punctated anterior prolongation; they are regular in size and distribution anteriorly in

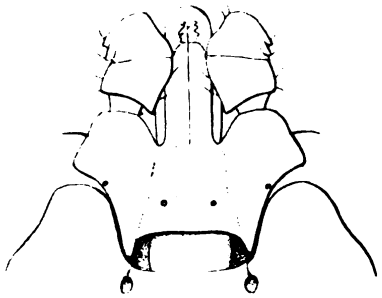


FIG. 24.—*Rhipicephalus longus*, ♂. Rostrum. × 45.

the space corresponding to the female scutum, and they exist with the same appearances in the posterior part of the scutum, but they become scanty and finer in the neighbourhood of the marginal grooves and their punctated anterior prolongations; the marginal border is smooth save in the scapular angles which are hollowed by six to eight larger punctations; posterior festoons longer than they are wide, almost smooth, followed by short abdominal festoons. *Eyes* flat, yellowish, of medium size, marginal with a coarse tangential punctation at the internal edge. *Ventral surface* reddish brown, with a few short hairs. *Anus* at about the middle of the length of the adanal shields; adanal shields long, semilunar (the inner edge concave, the external and the posterior convex) with fairly heavy punctations on their surface; external shields replaced by a prominent non-chitinous fold; festoons subrectangular, sharply defined; no caudal prolonga-

tion. Peritremes whitish, broad, comma-shaped, with point curved towards the dorsal surface. *Rostrum*, length 0·7 mm., dorsal base more than twice as broad as it is long, lateral angles very prominent at about the anterior third of the length, posterior angles quite prominent. Hypostome slightly spatulated, with six rows of teeth. Palps hardly longer than they are broad, flattened dorsally with the second segment a little longer than the third and shortened in a blunt point posteriorly at its inner border. *Legs* relatively strong. Coxae with long scattered hairs; first with anterior summit not prolonged nor visible on the dorsal surface, has two very long spines;

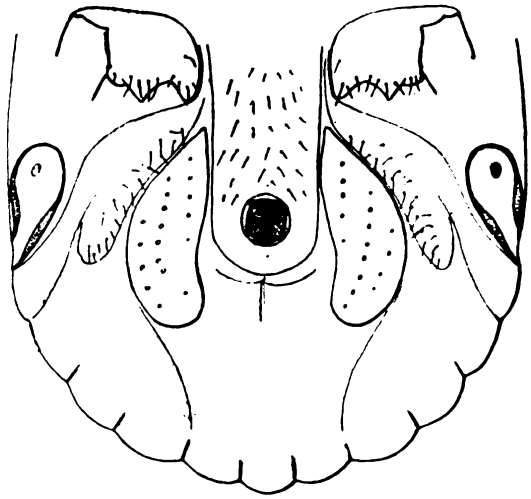


FIG. 25. *Rhipicephalus longus*, ♂. Ventral surface of posterior extremity. × 33.

second, third and fourth with posterior border incurved to form two short spines, the inner being wide, flat and blunt, the outer narrow and sharp. Tarsi of medium length with two terminal successive spurs; caruncules medium size.

FEMALE unknown.

Locality:—Diagnosis established from a male taken on a bovine at Kasongo (April).

Rhipicephalus duttoni and *R. longus* are placed in the following manner in the table of differentiation of males of the various species of *Rhipicephalus*.

1. (Eyes flat.—2.
 (Eyes prominent.

2. (Marginal groove well marked. 3.
 (Marginal groove absent.

3. (Dorsal scutum uniformly brown. 4.
 (Dorsal scutum black and white. *R. pulchellus.*

4. (Adanal shields not prolonged to a point (posterior border straight or convex).—5.
 (Adanal shields prolonged in one or two points (posterior border concave). —12.

5. (Posterior border of the body not prolonged nor furnished with a caudal prolongation. 6.
 (Posterior border of the body furnished with three prolongations.—*R. supertritus.*

6. (Adanal shields triangular or subtriangular (internal edge straight or slightly concave).— 7.
 (Adanal shields sickle-shaped (their inner border very concave, the two others forming a single regular curve).—*R. haemaphysaloides.*

7. (Marginal groove deep, long, commencing near the eyes. —8.
 (Marginal groove superficial, short, commencing at the middle of the length of the body.—*R. ziemanni.*

8. (Dorsal scutum with scattered punctations.— 9.
 (Dorsal scutum with numerous, serried, punctations.

9. (Dorsal scutum with unequal, very evident, irregularly distributed punctations.—10.
 (Dorsal scutum with large, equal punctations arranged in longitudinal lines with or without additional fine and hardly visible punctations.—*R. simus.*

10. (Coxae I, with or without a short prolongation in front, not visible from the dorsal surface.—11.
 (Coxae I, with a long anterior prolongation, visible from the dorsal surface.—*R. appendiculatus.*

11. Dorsal scutum with mixed punctation, regularly distributed. *R. sanguineus.*

Dorsal scutum with medium-sized uniform punctations between the marginal grooves, rare in their neighbourhood, none on the edge, large ones on the scapular angles. *R. longus.*

12. Anal shields with a single (or principal) internal point. - 13.

Anal shields with an external point.—*R. lunulatus.*

13. Coxae I, without visible prolongations from the dorsal surface ; adanal shields with two points.—*R. armatus.*

Coxae I, with prolongations visible from the dorsal surface ; adanal shields with one point.—*R. duttoni.*

ON SOME PARASITES IN THE MUSEUM OF THE SCHOOL OF TROPICAL MEDICINE, LIVERPOOL

ON SOME PARASITES IN THE MUSEUM OF THE SCHOOL OF TROPICAL MEDICINE, LIVERPOOL

BY

DR. A. LOOSS,

PROFESSOR OF PARASITOLOGY, SCHOOL OF MEDICINE, CAIRO

WITH A CONTRIBUTION ON

A CASE OF DISTOMIASIS OF THE LIVER AND THE RECTUM

BY

EDWARD CUFFEY,

PHYSICIAN TO THE BRITISH HOSPITAL, PORT SAID

During a visit I recently paid to the School of Tropical Medicine in Liverpool Dr. STEPHENS showed me a number of parasites which had been sent to the Museum of the School from various places. Unfortunately, some of the bottles bore no indication whatever as to the locality from which the specimens were obtained; others were labelled, but an inspection of their contents with the naked eye or a pocket lens raised within me strong doubts as to the correctness of the respective identifications. To make things sure Dr. Stephens kindly agreed that I should take with me, of those forms of which several were present, a few specimens for a more careful examination and possible identification of the species. The results of this investigation are given in the following pages.

Fasciolopsis buski (LANK.), 1857.

About a dozen specimens from Hong-Kong in a jar labelled "Distoma crassum." Rather large and fleshy, apparently some what contracted worms of about 30 mm. in length, 13 to 16 mm. in breadth and nearly 4 mm. in maximum thickness. Body moderately elongated, rather regularly oval, its surface marked with numerous fine transverse ridges brought about by contraction. Both suckers very close to the anterior end of the body; opening of the anterior sucker

minute and completely ventral, opening of the posterior sucker very wide, transversely oval, about 2 mm. in its largest diameter. The same is the average distance between the centres of the two suckers. In some cases the part of the body containing the anterior sucker was found separated from the rest in the shape of a minute but quite distinct cone resembling that which characterises the members of the genus *Fasciola*. Immediately in front of the ventral sucker there is a transverse slit indicating the position of the genital aperture. All round the margin of the body, the colour of the worms is considerably duller than in the middle and in most cases this duller tint projects twice on each side in an angular fashion into the lighter central area, this latter thereby assuming the shape of three successive but incompletely separated discs. All these characters discernible by naked eye inspection suggested *Fasciolopsis buski* (LANK.). A comparison of the internal organisation--so far as this could be made out from cleared entire individuals did not reveal any noticeable difference from the description recently given of the species by ODHNER (1902). The determination *Distoma crassum* is therefore correct, for the name *Distoma crassum* (COBB), 1877, is only a synonym of *Fasciolopsis buski* (LANK), 1857.

Distoma pancreaticum (JANSON).

A bottle from Hong-Kong, without label, contained a number of medium-sized flukes which in their outward appearance recalled the pancreatic fluke of ruminating animals in Eastern Asia. On closer inspection it could be seen even with the naked eye that there were two varieties present in this material, a larger form with very wide and projecting suckers and a somewhat smaller in which the suckers were well developed also, but much smaller and less prominent. Both varieties were represented by evidently completely mature individuals; there were, in addition, numerous young specimens of different size of the second variety, but no immature specimens of the first. Two specimens of the first variety and four adults of the small kindly given to me by Dr. STEPHENS form the basis of the present description.

After clearing, the chief points of their internal organisation could fairly well be made out in entire specimens; the details observed were in full agreement with the descriptions given of the *Distoma*

pancreaticum by the earlier writers. There were, however, between the two varieties in addition to the differences mentioned above in the size of the suckers several other slight internal discrepancies which leave no doubt that the two forms must be considered as two distinct species. Both are closely allied to each other and members of the same natural genus. The generic name hitherto usually adopted for *Distoma pancreaticum* is *Dicrocoelium*; indeed, the principal features of the anatomical structure are the same in *Distoma pancreaticum* and *Dicrocoelium lanceatum* (ST. and HASS.), the type species of the genus. But besides this similarity in the main structure there are also certain differences which to my mind forbid placing *Distoma pancreaticum* in the genus *Dicrocoelium itself*, that is to say as it is represented by its type *lanceatum*. One of the most conspicuous of these differences lies in the thickness and the considerable breadth of the body which latter allows the testicles to take a distinctly lateral position at about the same level, whereas in *Dicrocoelium* sensu strictiori, owing to the narrow spindle-shaped outline of the body, the testicles are placed obliquely one behind the other. In *Lyperosomum* which is also closely related to *Dicrocoelium* the two cross diameters are still more reduced, the members of this genus presenting to the observer an almost filiform body of about equal breadth and thickness in which the genital glands are found in a straight line one behind the other. In addition to the peculiar shape of their body the two *Distoma pancreaticum* differ from *Dicrocoelium* by the more complicated structure of their excretory vesicle and the stronger development of their copulatory organs. On the whole, therefore, there is sufficient reason for creating a new genus for the two forms under discussion.

The question as to the correct denomination of the *species* was somewhat difficult to decide. *Distoma pancreaticum* was discovered in Japan, and became first known in Europe during the Paris Exhibition in 1889, where the Agricultural School of Komaba exhibited a series of parasites among which there was a "*Distoma pancreaticum* from the pancreatic duct of sheep," and a "*Distoma pancreaticum* var. from the pancreas of sheep." The new parasite was referred to in subsequent years by RAILLIET (1890) and JANSON (1893 and 1895); the respective papers deal with the anatomical structure and the pathological significance of the parasite, they emphasise its

resemblance to *Dicrocoelium lanceatum* but do not -so far at least as I can personally consult them at present—contain any decisive statement from which one or the other of the two Hong-Kong species mentioned above might be recognised. In 1897, GIARD and BILLET, apparently ignorant of the existence of the *Distoma pancreaticum*, described a ***Distoma coelomaticum*** which the latter author had found plentifully in the pancreas of an ox killed at the slaughter-house of Cao-Bang (Tonkin); there had at first been some mistake as to the habitat of the parasite -which led to the denomination *coelomaticum* -but this mistake was corrected afterwards upon a suggestion of RAILLIET. The details given in GIARD and BILLET'S description are sufficient to show that *Distoma coelomaticum*, *also* structurally resembles *Dicrocoelium lanceatum* and *Distoma pancreaticum*, but there is again no statement which would be indicative of one of our two species in contradistinction from the other.

The latest description of *Distoma pancreaticum* is given by RAILLIET and MAROTEL in 1898. The specimens upon which their paper is based were collected by Dr. GOMY, a French veterinary surgeon, from the pancreatic ducts of cattle and Indo-Chinese buffaloes in Saigoon (Cochin-China), a locality therefore which is not so far distant from the place at which GIARD and BILLET'S material was taken. This time, the description is more complete; the worms are said to measure from 7 to 10 mm. in length and from 4 to 4·5 mm. in breadth; their suckers are of about equal size, the posterior of 0·75 to 0·95 mm. in diameter being only slightly larger than the anterior which presents a diameter of 0·7 to 0·9 mm. The distance between the two suckers varies from 1·3 to 2·4 mm., that is to say, it amounts to about a quarter of the total length of the body. As will be more fully seen hereafter, all these measurements apply very well to the smaller of the two Hong-Kong species and among the antomical details given by the authors there is none moreover which would not quite well fit in with its organisation. I thus think it perfectly safe to assume that this smaller Hong-Kong species is the same as the *Distoma pancreaticum* described by RAILLIET and MAROTEL in 1898 from Saigoon cattle. But RAILLIET in the same paper (p. 32) also states that he has satisfied himself *de visu* of the specific identity of this Cochin-Chinese pancreatic fluke with the parasite found in Tonkin by BILLET. This latter parasite had first

been described under the name of *Distoma coelomaticum* and the question now arises: is this *Distoma coelomaticum* of Indo-China really the same form as the *Distoma pancreaticum* of Japan? If this were so, then the smaller Hong-Kong form would have to be identified as the genuine *Distoma pancreaticum*, the name *coelomaticum* would have to be definitely dropped as a synonym and a new specific name would have to be given to the larger Hong-Kong species with the strongly developed suckers.

As I have already pointed out, the data given in the printed description of the original Japanese pancreatic fluke are not sufficient to decide the question. I possess, however, in my collection a number of specimens of *Distoma pancreaticum* which I owe to the kindness of Professor JANSON, of Tokio. They were collected from the pancreatic ducts of cattle in Japan and I think there can not be any reasonable objection to the assumption that they are specifically the original Japanese *Distoma pancreaticum*. It is true that the first specimens of this species exhibited in Paris were labelled as collected from sheep, but I am not inclined to attribute a great importance to this difference in the host, because it is a well-known fact that many parasites of cattle and buffaloes occur not only in sheep but in other ruminating animals also. A comparison of this Japanese material has now shown, first, that there is only *one* species present in it and, second, that this species is identical with the larger species of the Hong-Kong material. On the ground of these facts I feel justified in considering this latter species as the genuine *Distoma pancreaticum* of JANSON; for the smaller species I readopt the name *coelomaticum* of GIARD and BILLET, the suppression of which was, in the sense of the nomenclatural rules, a mistake based upon an erroneous identification of the species.

Eurytrema, nov. gen., *Dicrocoeliidarum*,

Differs from *Dicrocoelium* especially by the following particulars: Body rather thick and considerably broadened, with the exception of the hindmost part which retains its original shape and appears as a small triangular appendage sharply set off from the rest of the body; it is repeatedly referred to in the descriptions of the earlier authors as resembling in outline the cephalic part of *Fasciola hepatica*. Suckers very large and prominent, the oral sucker completely ventral and

surmounted by the anterior margin of the body. Excretory vesicle composed of a median stem which divides into two at about the middle of the total length, the transverse branches, in the neighbourhood of the intestinal caeca, dividing again each into an anterior and a posterior branch which run for the greater part outside of the intestinal caeca, and terminate in the neighbourhood of the anterior and posterior extremities of the body. Testicles lateral, at about the same level but far apart from one another. Copulatory organs well developed; cirrus pouch thick, almost cylindrical, containing a long but rather thin seminal vesicle and an equally long and thin ejaculatory duct which both describe a number of coils within the pouch. Vagina corresponds in length and arrangement to the ejaculatory duct. Structure of the remaining organs as in *Dicrocoelium*.

Type: *Eurytrema pancreaticum* (JANSON) 1889.

Eurytrema pancreaticum (JANSON),

(nec *Dicrocoelium pancreaticum*, RAILLIET and MAROTEL, 1898).

The two Hong-Kong specimens at my disposal are fairly well extended and measure 13 and 14 mm. in length; their maximum breadth of 6·5 and 7 mm. is reached immediately behind the ventral sucker. Thence the body narrows more quickly towards the anterior extremity than posteriorly but ends rather truncatedly owing to the large size of the oral sucker above which the margin of the body projects in the shape of a thick lip. The small tongue-like appendage of the posterior end is very conspicuous. The thickness amounts to almost 2 mm. in the median line of the anterior half, but gradually decreases towards the edge. The Japanese specimens are somewhat more contracted and thicker throughout, varying in length between 9·5 and 16 mm., in breadth between 5·5 and 8·5 mm.—in such a way that the longer specimens are generally less broad and vice versa. There are, however, also two apparently mature individuals 9·5 and 10 mm. long, but only 5·5 mm. broad. Skin, thin, without armature. Suckers very large and prominent, the anterior one with an average diameter of 2·1 mm., noticeably larger than the ventral which presents a diameter of 1·45 to 1·65 mm., but is in some specimens found more or less everted reaching thereby a size of 1·9 mm. In the two smallest specimens mentioned above the ratio of the suckers is 2·1 to 1·4 and 1·8 to 1·3 mm. respectively. They are, therefore, even in these

smallest specimens considerably larger than in the *Distoma coelomaticum* of the same size. The ventral sucker is always found near the end of the anterior half of the body; the distance between the centres of the two organs varying in the larger specimens from 4·5 to 5·7 mm. and being in the two small specimens 3·9 and 3·3 mm., that is to say it amounts to at least a third and is usually but little less than half of the total length. The genital opening has its position about halfway between the two suckers and is sometimes found slightly raised above the level of its surrounding. In one of the specimens the penis is seen everted, apparently to its full length, for the larger part of the seminal vesicle is located in it; it represents an organ 3·4 mm. in length and at its base 0·37, at the free end 0·24 mm. wide. Its surface is smooth.

The internal anatomy agrees with the descriptions given by the earlier writers. The small pharynx of 0·47 mm. length and 0·54 mm. width is usually so much displaced dorsally that it is not visible from the ventral aspect; behind it there follows a short oesophagus which, as a rule, runs straight down towards the ventral side and may thus more or less entirely disappear from view. The intestinal caeca of proportionally insignificant calibre quickly diverge and follow a zigzag course which becomes especially pronounced behind the testicles. Their blind terminations lie about as far as the length of the tongue-like end-portion of the body in front of the base of that portion and usually not quite on the same level, the left branch being as a rule (but not always) the shorter. The full extent of the excretory vesicle could not be made out in any of the specimens at my disposal, owing to the thickness of the body and the strong development of the uterine coils. The main parts described in the generic diagnosis are usually seen and followed without much difficulty; but, in addition to them, one may observe in some places isolated portions of similar tubes which seem to be ramifications of the anterior and posterior lateral branches. There are two running forwards on either side of the ventral sucker, and two running backwards and towards the median line amidst the uterine coils in the posterior half of the body Although they appear as if they were parts of the excretory vesicle, I cannot tell with certainty whether or not they really belong to that organ. The terminations of the main anterior branches of the vesicle lie close to the oral sucker at about the level of its centre, those of the

posterior branches at, or slightly in front of, the two notches which separate the terminal triangular tongue from the rest of the body.

The position of the genital aperture has been given above; it lies always a little behind the bifurcation of the intestine. The thick muscular cirrus pouch, usually somewhat curved in the shape of a sausage, presents a length of 2·2 to 3 mm. and an average width of 0·6 mm. Its posterior end rests upon the anterior slope of the ventral sucker, but there are some variations in its length in connection with the contraction of the body and the fulness of the seminal vesicle. This latter occupies the posterior half of the pouch, describing, within it, a proportionally large number of small coils. The pars prostatica is short and the number of glandular cells surrounding it limited. The ejaculatory duct is at first rather narrow, but widens somewhat towards its free end; its coils are proportionally numerous also. The dimensions of the evaginated penis have been given above. The fairly large testicles are normally found at the same level somewhat behind the ventral sucker, but may in strongly contracted specimens be more or less completely shifted to its sides. They have in the greater number of my specimens a distinctly lobed shape, but exceptionally the lobes may be so short and broad that the glands appear to possess a merely notched outline. The number of lobes (or notches) seems to be constantly four for one (usually the left) and five for the other testicle. These conditions are inverted in the cases of "sexual amphitypia" which are not unfrequently met with in this as well as in the following species.

The small ovary is usually found a short distance behind the testicles, on the left—in cases of sexual amphitypia on the right—side of the body. It has in all the specimens with lobed testicles a distinctly lobed shape also, the number of lobes being three. In those specimens, on the other hand, in which the testicles show the notched outline only, the ovary, too, loses its lobed outline more or less and appears compact with some irregular bulgings of its surface. The oviduct starts from its near side and almost immediately enters into a well-developed shell-gland which in cleared specimens appears as an opaque sharply outlined body by the side of the ovary and close to the ventral surface. There is also a fairly long LAURER'S canal of an average diameter of 0·04 mm. which opens to the outside in front of the ovary. It carries a small seminal receptacle which I have never been able

to discover in entire specimens, but clearly saw in a series of sections. The receptacle represents in the sectioned specimen a globular body of about the same size as the ovary (0·4 mm. in diam.) and lying close to its dorsal surface. It communicates with the LAURER'S canal by a distinct thin duct, but does not contain many spermatozoa, a fact which renders the seminal receptacle very transparent in unstained specimens. This insufficient filling in combination with the particular position of the seminal receptacle is obviously the reason why it escapes observation in entire worms.

The yolk-glands are only little developed; they are composed of rather numerous acini of a slender club shape collected into more or less distinct groups, the number of which seems to vary between ten and twelve. In extended specimens the groups lie in a nearly straight line one behind the other and are fairly well discernible; in contracted individuals they become more closely packed together and some of them—especially those in the middle—may be shifted laterally in such a way that for a certain distance there appear to be two irregularly alternating rows of groups. In this latter case some of the follicles are pushed below the intestinal caeca, otherwise, the yolk-glands lie as a whole between these and the body margin, usually the gland on the side of the ovary somewhat farther in front than that of the opposite side. Their extent varies a little individually and with the contraction of the body. In the greater number of specimens they commence somewhere at the outer border of the testicles and their posterior ends lie halfway between the posterior border of the ventral sucker and the base of the tongue-like end-portion of the body, but may in strongly contracted specimens also be found further forward. The transverse yolk-ducts originating, on each side, by the union of an anterior and a posterior tributary arise from the glands at about their middle; they join near the ovary (the left transverse yolk-duct being thus shorter than the right) and then enter the complex of the shell-gland from behind. There is no distinct widening of the unpaired part of the yolk-duct into a yolk reservoir. The coils of the uterus are very numerous but do not, as a rule, appear to the observer in the shape of regular loops, but rather as loose tubes branching and sometimes anastomising in an irregular manner. Occasionally only, i.e. in individuals in a certain state of contraction, one may perceive the picture which is so characteristic of the uterus of *Dicrocoelium*

lanceatum and in which the descending and ascending stems of the uterus cover each other in such a way as to appear as a straight main stem from which lateral ramifications are branching off. As far as the branches of the intestine extend backwards, it is only exceptional to see one or the other uterine coil cross them towards the margin of the body; behind their blind ends the coils extend much nearer to the margin and the terminal small triangular appendage is, in perfectly mature specimens, almost completely filled with them. Anteriorly the coils are limited by the testicles and the ventral sucker; the uterus then passes between the latter and the right testicle (i.e. the testicle opposite the side of the ovary) and in front of this still forms some small convolutions before entering into the long and thin, but rather muscular vagina which accompanies the cirrus pouch while describing a number of narrow coils.

The eggs have a striking likeness to those of *Dicrocoelium lanceatum*; they are slightly larger than these showing, on an average, a length of 0·05 mm. and a width of 0·034 mm.—but apart from that, they present the same oval shape and the same thick shell of deep brown colour as these and contain, in addition, a miracidium which, when fully developed, shows the same two enigmatical strongly refractive bodies which are peculiar to miracidia of *Dicrocoelium lanceatum*.

Eurytrema coelomaticum (GIARD and BILLET) 1892.

(Synonym: *Dicrocoelium pancreaticum* RAILLIET and MAROTEL, 1898).

The largest specimens at my disposal attain a length of 10 mm., and a maximum breadth of 5·5 mm.; younger (immature) specimens measure 7·5 by 4, 7·5 by 3·5 mm., &c. The outline of the body is almost regularly oval, and there is no marked narrowing towards the anterior extremity. The triangular terminal portion ("languette triangulaire" of RAILLIET) is fairly conspicuous. The maximum thickness of the body does not seem to exceed 1 mm. In the four large (mature) specimens which I have for examination the oral suckers present diameters of 0·95, 0·88, 0·85 and 0·83 mm., the ventral suckers diameters of 0·98, 0·91, 0·85 and 0·83 mm. respectively. These

measurements agree fairly well with those given by RAILLIET who, in the Saigoon specimens, found the oral sucker to vary in diameter from 0·75 to 0·93 mm., the ventral sucker from 0·7 to 0·9 mm. The distance between the centres of the two organs which RAILLIET gives as 1·3 to 2·4 mm. is in the four above mentioned specimens, 2·5, 2·97, 2·7 and 2·72 mm.; the suckers are therefore, in *Eurytrema coelomaticum*, proportionately much nearer to each other than they are in *E. pancreaticum*, for the ventral sucker lies in the former species with the greater part of its mass still in the anterior third of the body and the distance between the two always remains *less* than a third of the total length. The size and relative position of the suckers represent, apart from the general shape of the body, the most conspicuous of the specific characters of *Eurytrema coelomaticum*.

The internal anatomy has been so fully described by RAILLIET and MAROTEL that I have nothing of importance to add; it is moreover essentially the same as that of *E. pancreaticum*, as may be seen by a comparison of the data given above with the description of the French authors. There are nevertheless the following differences in detail which I am inclined to consider as of specific value, although some of them are not absolutely constant. In *E. coelomaticum*, the genital glands never present the lobed appearance of those of *E. pancreaticum*, but always show a compact shape with irregular bulgings of their surface instead of lobes; the apparent rule that one testicle has four the other five bulgings seems however to be preserved here too. The yolk-glands which, in *E. pancreaticum*, were composed of ten to twelve groups of follicles, show in *E. coelomaticum*, only six to eight; the glands are therefore as a whole shorter than in the preceding species and they also begin somewhat farther backwards, viz. at the same level with the *posterior* margin of the testicles. In spite of that, their posterior terminations appear much more distant from the end of the body than in *E. pancreaticum*, a fact due to the considerable difference in the relative position of ventral sucker and genital glands in the two species. In *E. coelomaticum*, finally, it is not so uncommon to see one or the other uterine coil pass beyond the intestinal branches laterally.

The eggs, in shape, aspect and structure of the miracidium, closely resemble those of *E. pancreaticum*, but are somewhat smaller, namely

0·042 to 0·046 mm. long and 0·023 to 0·027 mm. wide. This size, by the way, is the same as that of the eggs of *Dicroc. lanceatum.*

Gastrodiscus secundus n. sp.

A small jar containing about a dozen specimens and a label on which there was written in pencil: "Gastrodiscus Sonsinonis Cobb. from mule." They were collected by GILES in Assam, who states that they are also found in equines throughout India. In their external appearance they absolutely resembled the African *Gastrodiscus aegyptiacus* (COBBOLD) 1877 (= *Diplostoma aegyptiacum* COBBOLD 1877, = *Gastrodiscus polymastos* LEUCKART 1877, = *Gastrodiscus sonsinoi* COBBOLD 1877), with the sole difference that all of them were noticeably *smaller*. Three specimens at present at my disposal show a length of 7, 8 and 8 mm., and a maximum breadth of 4·5, 5 and 5 mm.; the corresponding measurements in *Gastrodiscus aegyptiacus* being 10 to 13 mm. and 7 to 9 mm.

The body shows the characteristic division into two parts: the cylindrical or slightly depressed cephalic portion, and the broad oval disc, at the end of which the second sucker is placed. In the specimens under discussion the former is about 1·35 mm. long and at its base 1·6 mm broad. Near its top, there appears on the ventral surface the mouth opening, about 0·37 mm. wide. In its neighbourhood the cuticle shows a large number of small circular or oval thickenings which project externally in the shape of small tubercles. They are apparently tactile papillae similar to those found round the mouth opening in allied forms. Behind the mouth the ventral surface of the cephalic cone is slightly excavated along the median line. The abdominal disc is apparently much contracted, for its free edge is rolled inwards to such an extent that the ventral surface and the terminal disc are visible in part only. The ventral surface is, so far as it lies open to the observer, densely covered with the well-known "papillae," or "pseudo-suckers" bearing on their summits small slit-like openings. Some of these organs are everted and, in this state, represent small knobs projecting somewhat over their retracted neighbours. On inspecting the worms with a magnifying glass, a small circular hole surrounded by a somewhat raised brim becomes visible in the midventral line about 1·35 to 1·5 mm. distant from the base of the cephalic cone. It was present at the same place in all the

specimens and therefore a normal structure, probably the genital aperture. If the latter was true then the worms could not belong to the species *aegyptiacus*, for in this, the genital aperture is placed immediately behind the cephalic cone, at the very base of the projecting edge of the disc.

A close microscopical examination of the three specimens kindly given to me by Dr. STEPHENS has confirmed this presumption; the parasites represent a new and at the same time a true member of the genus *Gastrodiscus*. It appears superfluous to give a description of its internal structure here, for this would mean nothing but a second description of the structure of *Gastrodiscus aegyptiacus*. It may therefore suffice to point out the differences between the two species. They are chiefly given in the position of the genital aperture and the extension of the yolk-glands. The former has already been mentioned; it is a circular or transversely oval opening about 0·3 mm. wide, and leads into a genital atrium of 0·3 mm. in cross diameter, the muscular wall of which is of considerable thickness and much more conspicuous than in the African species. The floor of the atrium is again raised in the shape of a conical papilla on the top of which the male and female genital ducts open close by one another in minute pores. The yolk-glands show a richer development than in *Gastrod. aegyptiacus*. In this species, it will be remembered, they are located in the ventral half of the body, between the intestinal caeca and the free margin of the disc, and it is rather exceptional to find some folliculi extending below the caeca into the space between these and the median line of the body. In *Gastrodiscus secundus*, the yolk-glands trespass on the intestinal caeca in such a manner that their loosely grouped follicles occupy, in the ventral half of the body, all the space left free by the other organs. A minor difference is afforded by the size of the suckers. The oral sucker which, in *Gastrod. aegyptiacus*, presents a cross diameter of 0·8 mm shows, in *G. secundus*, a diameter varying, according to the size of the specimens, from 0·55 to 0·65 mm.; the posterior sucker, in *G. aegyptiacus* about 2 mm. wide, in *G. secundus* scarcely reaches 1·2 mm. in size. The uterus, in the body of the new species, principally takes the same course between the organs as in *G. aegyptiacus*, but, although it is, in the two larger individuals at my disposal, thickly filled with ova, it seems that it does not form so many secondary loops as in the older species. The ova

which, in the latter, show a length of 0·17 to 0·19 mm. and a width of 0·11 mm., are, in *G. secundus*, somewhat smaller namely 0·15 to 0·16 mm. long and 0·09 to 0·1 mm. wide.

Opisthorchis sinensis (COBB.) 1875.

Several worms of a rusty yellow colour contained in a jar of unknown origin and labelled "*Distoma sinense* COBBOLD" struck me at once by the fact that they were considerably larger and stouter than any others I had seen of the species up to that time. I therefore had no hesitation in expressing the opinion that these worms could not be the true *Opisthorchis sinensis*. Since the number of specimens was limited, I took two specimens only with me, an unhurt one and another which was broken into two pieces. About a fortnight after my visit to Liverpool I paid a visit to the "Institut für Schiffs-und Tropenkrankheiten" in Hamburg. Dr. FÜLLEBORN very kindly showed me round and, during an inspection of the collection of human parasites, I was rather surprised to recognise under the designation "*Opisthorchis sinensis*" the same large form which I had seen a short time previously in Liverpool. There were about a dozen individuals, part of them kept in spirit, others mounted as microscopical preparations. They had all been collected from one case, a Chinaman who had died in the Seamen's Hospital in Hamburg. Besides these large specimens, there was one microsopical preparation of an ordinary small specimen such as those I had alone seen before. This specimen was *not* derived from the case from which the large worms had been obtained. Dr. FÜLLEBORN was kind enough to confide this whole material to me for comparison with the Liverpool specimens. On my return to Egypt, four weeks later, I learned that a short time previously, a great number of "liver flukes" had been found at the post-mortem examination of a Chinaman who had died in the British Hospital of Port Said. Suspecting a case of Opisthorchiosis I wrote to Dr. E. H. ROSS, Public Health Officer of the Town and Port of Port Said, to ask for some specimens of the worms. Thereupon, Dr. CUFFEY, who had had the patient under his care and had made the post-mortem examination, most obligingly placed at my disposal all the preserved material of the case. He was also kind enough to write a short abstract of it which I insert in full here

because of some interesting particulars it contains.* The preserved material sent to me consisted of about 200 loose worms, and many others were still seen densely packed in the bile-ducts of some preserved pieces of liver. Among the loose worms there was again not a single specimen of the ordinary small variety, but all were the large form. I had thus, within a very short lapse of time, thrice come across a variety of the Chinese liver-fluke which I had never seen before, and in all these three cases this variety had been present alone, quite unmixed with individuals of the usual smaller variety.

I had not the least doubt that this new form would prove to be an independent species and that it would be easy to delimit it anatomically from its smaller congener. I was therefore somewhat disappointed, when after a most minute and thorough comparison of the specimens with those I possessed of the small "*Distoma sinense*," I could not, apart from the size of the body and the ratio of the suckers, find any constant internal differential character between them. Even of the characters mentioned the last-named is by no means conspicuous, and I had almost made up my mind that the two varieties must be considered as one and the same species, when, on reconsidering all the factors involved, I came to the conclusion that

* A CASE OF DISTOMIASIS OF THE LIVER AND THE DUODENUM.

Too-ting, a Chinaman, aged 25, was admitted to the British Hospital, Port Said, on May 10th, 1906, suffering from Lumbar Abscess. He was landed from a ship proceeding from China to England. The abscess was pointing in the lumbar region, posteriorly, 2½ inches from both the middle line of the back and the crest of the ileum. It was opened and about 300 grms. of bile-stained pus were evacuated. The walls of the abscess were explored and the transverse process of one of the lumbar vertebrae was found to be necrosed. But no connection could be found between the abscess and the liver. The cavity was washed out and stitched up. Subsequently a drainage tube had to be inserted, for the pus collected again; but the man's condition rapidly improved, and the wound over the site of the abscess healed completely.

On June 21st, the man suddenly developed pneumothorax, affecting the lower part of the right side of the chest. As he was thought to be suffering from an abscess of the liver, this organ was explored again, but no pus was found. Oedema occurred over the lower part of the right side of the chest and still no pus could be found on exploration. The patient rapidly became worse, and died on June 25th.

Post Mortem.—The abscess in the lumbar region had healed completely. No reason for it could be found beyond the necrosed vertebra.

Liver: The liver was enlarged and contained numerous abscesses in both lobes. They varied in size from that of a small pea to a tangerine orange. On section the liver presented hundreds of liver flukes in the bile-ducts. The hepatic ducts, common bile duct, and gall-bladder all contained numbers of these flukes.

They were also found free in the duodenum, but not in the ileum.

There were no flukes in the abscesses.

No cause for the pneumothorax could be found, and no connection between it and the liver was apparent. All the other organs were normal.

(Signed) EDWARD CUFFEY,
Medical Officer, British Hospital,
Port Said.

such a ruling would be less defensible than the other alternative, viz. to declare them as separate species.

I am pretty certain that many of my fellow-helminthologists will think I am pushing things too far, and that what I am about to do here is nothing but "species-manufacturing." In order to meet this reproach as far as I can, I may be permitted to give my reasons somewhat in detail.

To begin with, it is a fact acknowledged by all observers who have dealt with this particular group of parasites (BRAUN, LUEHE, MUEHLING, LOOSS) that the members of the genus *Opisthorchis* are in many instances by no means easily to be distinguished. As one out of several examples I will mention the case of *Opisthorchis felineus* and *Opisthorchis geminus*, a case observed by myself. The former species is in Europe a rather common parasite in the liver of certain beasts of prey, and especially in cats, and is in eastern Europe even a not uncommon occasional parasite of man. *Opisthorchis geminus* inhabits in Egypt the liver of certain birds (*Milvus aegyptius*, the common Egyptian kite, *Circus aeruginosus*, a glede, and *Anas boschas fera*, the wild duck). It resembles *Opisthorchis felineus* anatomically to such an extent that the two forms are hardly distinguishable from one another without knowledge of host and locality. Their internal organisation, therefore, would not furnish unobjectionable reasons for separating them specifically; nevertheless, they must be considered as specifically different on biological grounds —or else we must admit as true certain circumstances which are almost impossible to comprehend. Among the hosts of *Opisthorchis geminus* *Milvus aegyptius* is a true Egyptian who never leaves the country and thus must acquire his parasites *in* it. But if *he* does, it would be inconceivable why dogs, cats and even Man—hosts that in Egypt actually *share* other species of parasites—should not pick them up also and become infected as they do in Europe, *if* the parasite of the Egyptian birds and that of the European mammals were indeed one and the same organism. Never, however, have I in Egypt found any trace of an *Opisthorchis* form in canine or feline animals, wild or domestic; nor in Man, although I have purposely looked for them whenever there was an occasion; nor have the professors of pathology who within the last ten years have made the post-mortem examinations at Kasr-el-Aini Hospital noticed anything of the sort. It can therefore

be considered as certain that no *Opisthorchis* species occurs in mammals and Man in Egypt and the inference therefrom *must* I think be that the Avian *Opisthorchis* which is indigenous in this country cannot be the same species which inhabits mammals and Man in Europe—all the resemblance they show in their internal organisation notwithstanding.

The case of *Opisthorchis felineus* and *Opisthorchis geminus* is not the only one known; but this as well as the corresponding instances tend to show that, in the case of the two *Opisthorchis sinensis*-forms also, the internal similarity must not necessarily be a proof of their specific identity—the less so, as in the size of their bodies and the ratio of their suckers characters are given which practically allow a differentiation of the two forms.

A second point which seems to me very important is the fact that the two varieties of *Opisthorchis sinensis* not unfrequently occur quite separately. It has already been pointed out that the large variety was present alone in the three cases mentioned above; but the same fact may be inferred from some earlier cases on record. The parasite was discovered in 1874 by McCONNELL; the measurements given of the worms by the author are $\frac{7}{10}$ in. for the length and $\frac{1}{7}$ in. for the breadth. This would correspond to 17·8 and 3·63 mm. respectively and is, as may be mentioned here in anticipation, exactly the size of the specimens I at present possess of the large variety.* The author

* It is perhaps not out of place here to draw attention to the fact that the size of preserved specimens of parasitic worms is within certain limits influenced by the methods of preservation on the one hand, and by their own condition at the moment of preservation on the other. If specimens are collected from a living or recently killed host, they are still alive; if they remain in the organs after the death of their host they will die, too, after a shorter or longer time; the same thing happens when living parasites are brought into water, which is very harmful to them and destroys certain species very quickly. Before death they stretch themselves to their full size, and the commencing decomposition even makes them swell somewhat. Living parasites, when preserved immediately after removal from their natural habitat, always react by a contraction, which is the stronger the slower the preserving reagent acts upon them. Specimens which have died—in water or in the organs of their hosts (as is usually the case in post-mortem examinations made some time after death and in warm climates)—do no longer contract, but may shrink more or less according to the dehydrating power of the preserving fluid. All these factors, therefore, have their influence upon the dimensions the specimens exhibit after preservation, and it is necessary to remember this fact when the size of a certain species is used as one of its distinctive characters. However, the changes due to preservation always keep within certain narrow limits, and can even be fixed by actual observation. The parasites of the Port Said case were obviously dead when they came into contact with the preserving fluid, for they are not contracted. The fluid used had been formaline, which does not dehydrate; thus, the specimens show an average length of 16 to 18 mm., which is about their natural length at full extension. Some of them were afterwards most carefully and gradually transferred into alcohol; when the strength of this had reached 90 per cent. the worms had shrunk to a length of 14 to 16 mm., but could be made to very nearly assume their former length again by placing them back into formaline.

adds that the breadth is very constant in all the specimens, whereas the length presents slight variations from $\frac{6}{10}$ in. to $\frac{8}{10}$ in. (= about 15·3 to 20 mm.). In 1877, the parasite was recorded by MACGREGOR as the possible cause of a peculiar form of paralytic disease, of which he had seen three fatal out of eight cases. The observations were made in Port Louis, Mauritius, but all the patients were again Chinamen. I have at present no access to MACGREGOR'S original article, but the parasites found and described by him were subsequently examined by COBBOLD and recognised as identical with those of MCCONNELL. In 1878 MCCONNELL himself describes a second case observed by him in Calcutta; "the subject was, curiously enough, again a Chinaman," and the distomata found "were quite identical with those originally found." Comparing these earliest observations we see that in all cases the bearers had been Chinamen, and that the parasites must have been of about the same size for no allusion is made to the existence among them of any noticeably smaller individuals.

In 1883, the presence of flukes in the liver of Man was reported from Japan, first in a paper written in Japanese ("Observations on Distomes in the Lung and Liver") by KIYONO, NAKAHAMA, SUGA and YAMAGATA, acting physicians in the hospital of Okayama, and a short time later by BAELZ in a paper "Ueber einige neue Parasiten des Menschen." Unfortunately, I have at present no access to this paper which is one of great importance for the question I am now discussing; thus I can here refer to its contents so far only as they are reproduced by later writers. According to these, BAELZ recognises two distinct species of liver-flukes, a *Distoma hepatis endemicum* sive *perniciosum*, and a *Distoma hepatis innocuum*. They are distinguished from one another (see LEUCKART, 1876, p. 338), first and foremost by their quite different size, *Dist. innocuum* attaining a length up to 20 mm., whereas *Dist. endemicum* does not exceed 8 to 11 mm. Of the other differential characters given by BAELZ the following seem to me to be worth mentioning: *Dist. innocuum* has (1) a uterus of lighter colour but larger volume, (2) slightly larger ova than *Dist. endemicum*, and (3) possesses in its excretory apparatus and body-parenchyma numerous black granules. The exact measurements of the ova are, according to BAELZ, in *Dist. innocuum* 0·021 to 0·036 mm. length by 0·018 to 0·002 mm. breadth, in *Dist. endemicum* 0·02 to 0·03 mm. length by 0·015 to 0·017 mm.

breadth. From the names given to the species it may be inferred that the small variety is very common in the country ("endemic") and harmful to its host ("pernicious"), presumably because of its occurrence in large numbers, whilst the larger is rarer, i.e. not present in large numbers and therefore comparatively harmless ("innocuous").

In subsequent years the Japanese liver-flukes were the object of repeated scientific investigation. It was soon recognised that they bore a close anatomical resemblance to the Chinese liver-fluke discovered by McCONNELL; as a matter of fact, all later authors are agreed in declaring them more or less definitely as identical with one another as well as with the *Distoma sinense*. A comparative study of the papers published by these authors, however, reveals an interesting fact, namely that so far as specimens of Japanese origin are concerned these authors, with one exception, do not seem to have ever again seen the large variety, i.e. the *Dist. innocuum* of BAELZ. Thus in 1886, IJIMA, when describing *Dist. endemicum*, gives the average length of the body as $11\frac{3}{4}$ mm., whilst the largest specimens measure 13, the smallest 8 mm. in length; the breadth is 2 to $2\frac{3}{4}$ mm. LEUCKART who named the *Distoma sinense* of COBBOLD *Distomum spathulatum*, examined part of BAELZ'S original material and specimens sent to him by IJIMA; he found the worms to vary in length from 10 to 13 mm. and in breadth from 2 to 3 mm. I saw this material myself at the time and do not remember having noticed any remarkable difference in size of the specimens. In 1900, KATSURADA published a very exhaustive paper based upon 76 post-mortem examinations in which altogether an enormous number of parasites were found; in 21 cases indeed their number exceeded 1,000 and in five there were between 2,000 and 5,000. It may be mentioned here in passing that KATSURADA for the first time states that in cases of strong infection the parasites also occur in the pancreatic ducts causing there sometimes no perceptible changes, but generally a dilatation of the ducts combined with a thickening of their walls; occasionally, one may even observe inflammatory infiltration and hypertrophy of the connective tissue in the neighbourhood which seems to lead to atrophy and degeneration of the glandular substance.

With regard to the question which interests us here, it is to be regretted that KATSURADA does not give exact measures of individual specimens, but averages only derived from measurements of a certain

number of worms found in separate cases. Thus, five individuals measured of one case gave an average of 11·0 mm. length by 2·50 breadth; 21 individuals of another case gave an average of 9·78 mm. length by 2·40 mm. breadth; and 35 individuals of a third case gave 14·06 mm. by 3·88 mm. Nothing need be said concerning the two first cases; in the third, however, it is almost certain that large individuals of 16 to 18 mm. length have been present, although the author does not mention it especially. KATSURADA explains the variations in the size of the worms by circumstances such as size of the host, number of parasites actually present at the same place, &c., and says that "in cases where few worms only inhabit a full-grown man many of more considerable size are found among them; whilst, on the contrary, in children, dogs and cats the worms are generally smaller."

This view, by the way, which was still very generally adopted not so many years ago, is not supported by actual observations. I have for several years paid special attention to this question, but all my experiences tend to show that every species of parasite has a size of its own. There *may* occasionally be individual specimens which trespass the normal size of their race more or less markedly (just as this occurs in Man and higher animals), but, as a rule, their size is very much the same, whether they live in a large or in a small host. A full-grown *Ascaris lumbricoides* is as large in a child of one year's age as it is in an old man of seventy; and the common liver-fluke is not smaller in a goat or a sheep than it is in a cow, nor is it, when full-grown, smaller in a narrow bile-duct than in a wide one. In the above mentioned case of *Distoma sinense* observed by Dr. CUFFEY, the worms thickly crowded the bile-ducts of medium calibre; nevertheless, not a single small specimen could be detected among the loose ones. The small *Distoma endemicum*, on the other hand, retains its specific size quite irrespective of the number in which the individuals may be present in a given host.

The observations so far spoken of had all been made on material collected in Japan. During the same time, however, the parasite had also been discovered in subjects coming from other parts of the Extreme Orient, such as Corea, Tonkin, and the island of Formosa. I do not possess the special literature on these earlier cases, but may refer the reader to an article by R. BLANCHARD, "Note sur quelques

vers parasites de l'homme (1891)" in which the necessary references are given. In this same article BLANCHARD also records three new cases from which the parasites obtained had been submitted to him for examination. The material consisted of two bottles containing each several hundreds of *Distoma sinense* which had been collected at the post-mortems made of two Annamite soldiers in the military hospital of Hanoï. A third bottle from Hanoï contained, in addition to a piece of liver, an extraordinary number of parasites; so many indeed that the author says that without the positive affirmation given it would have been hard for him to believe that they had actually been derived from one and the same autopsy. At another place (1901, p. 581), he says that one might estimate the number of these worms as being rather above ten thousand. The statements BLANCHARD makes with regard to the external aspect of the parasites are so interesting that I reproduce them in the original language (1891, p. 5). "Dans l'un des deux flacons reçus de Mr. le Dr. TREILLE (i.e. those mentioned above in the first place), tous les Distomes sont de grande taille et semblables entre eux. Dans le second flacon, ainsi que dans celui que j'ai reçu du Dr. LOYE (the third bottle), on remarque au contraire des Vers de deux sortes: les uns, d'aspect très foncé, longs de 14 millimètres, larges de 3 mm. 2; les autres, beaucoup plus clairs et mesurant seulement 8 millimètres sur 2 millimètres."

Thus, there was again one case in which the large form was present alone, whereas in the other two both were represented, but so clearly recognisable from each other that the author immediately goes on saying: "A première vue, on dirait qu'il s'agit de deux espèces distinctes . . ." The conclusions drawn by BLANCHARD from these observations will be discussed later on; on the present occasion, I wish to draw attention to the fact that the large specimens were "d'un aspect très foncé," or, as I interpret it, strongly pigmented, whilst the smaller specimens were "beaucoup plus clairs," that is to say without pigmentation. The size of the worms recorded by BLANCHARD is somewhat below the average for both varieties, very probably, therefore, they are somewhat contracted owing to the action of some stronger reagent.

Reviewing now what has been recorded on the preceding pages we have the following facts:—The original *Distoma sinense* of MCCONNELL and COBBOLD has up to now chiefly been found in

Chinese and is at present the only species of liver-fluke known from this race. It reaches an average size of 18 by 3·5 mm. and has a more or less opaque body owing to the presence within the body parenchyma of granules of a dark pigment. In Japan, similar flukes are also very common in the liver of Man and certain animals; but here they vary, especially in size, to such an extent that one of the first observers (BAELZ) felt justified in establishing two distinct species. One of them coincides in size with the *Distoma sinense* and also possesses the pigment granules in its body parenchyma. It is, in Japan, apparently not very common, for some observers subsequent to BAELZ have not come across it at all during their investigations, whereas BAELZ himself considered it as harmless—presumably because he never saw it in large numbers—and KATSURADA explains the large size of the specimens he saw in certain of his cases by the fact of few individuals only being present at the same place. The second species is markedly smaller, attaining an average size of 11 to 12 by 2 mm. only; it is not pigmented and appears to be very common in Japan as indicated by the name "endemicum" and, furthermore, by the fact that several observers saw this form alone during their researches. There are finally the three cases recorded by BLANCHARD; in one of them only large worms were present, in the two others, a large and a small variety were found, the former remarkable by its dull colour, the latter much lighter.

From these observations I cannot but draw the conclusion that in the two differently sized varieties of the *Distoma sinense* we actually have to deal with two independent forms. One of them (the larger) is indigenous on the Asiatic continent, especially China, but is occasionally also found in Japan (and probably the other islands of Eastern Asia), the other is indigenous in Japan and common also in Annam and Tonkin (possibly all along the sea-shore in the Far East).

This view differs from the conclusion arrived at by BLANCHARD. As quoted above, his first impression on seeing the two different sizes was that he had two distinct species before him; a close comparative examination, however, convinced him of the contrary. "La disposition anatomique est la même dans les deux cas; la structure et les dimensions de l'oeuf sont également indentiques. On constate simplement que les plus petits individus ont les vitellogènes moins

développés et l'utérus chargé d'oeufs en moins grand nombre. Cette observation démontre donc que les petits individus sont des jeunes, non encore parvenus à toute leur croissance ; elle explique en même temps leur aspect plus clair." I may confess that for a certain time I held a similar view ; but it cannot be upheld any longer for the following reason : Supposing that the small individuals were indeed young, i.e. in the beginning of the period of their sexual activity, one ought to expect that the large individuals would be without exception perfectly sexually mature and full of ova. But this is actually not the case. In the Hamburg material I have for examination there are two immature specimens, both with a uterus developed to its full length but containing, in the first specimen, only spermatozoa (the seminal receptacle containing nothing but a number of egg cells), and in the second specimen a number of loosely grouped ova which are still far from filling the coils as they do in ripe individuals. Nevertheless, these worms are unmistakeable representatives of the large species, for they measure, the first 13·5 by 2·5 mm., the second 12·5 by 2·75 mm. ; they show, in addition, the size of the suckers and the brown pigmentation distinctive of that form. One similar specimen was found in the material from the Port Said case. For all of them, the explanation alluded to above does not hold good ; they prove, on the contrary, that the large species when the period of its sexual maturity begins has already reached, or even exceeded, the maximum size of the small form. Unfortunately, the scant material I possess of the latter does not contain any young individuals ; but the fact just recorded suffices I think to show that the two forms cannot be one and the same species.

If they are different the question arises, whether they are to be considered as full species or as mere varieties. I suppose that many of my fellow-helminthologists, regarding, in the first place, the almost perfect similarity of the internal organisation, as compared to which the differences in size, &c., appear insignificant, would find the latter alternative quite sufficient to cover the facts, assuming perhaps so-called " local varieties." I unhesitatingly proclaim the former view as being more logical and more consistent. The chief character of the species, in contradistinction from the variety, lies in the constancy with which it preserves its specific peculiarities. The variety is inconstant in a larger or lesser degree and alternates with typical specimens of the

species. A similarity in the internal organisation, even if it is very close, is not decisive for a variety, for otherwise it would be a simple logical necessity to consider for instance the cat as a mere small variety of, let us say, the leopard or the rat as a large variety of the mouse. Both cat and leopard, and rat and mouse, are internally almost absolutely alike and yet they are no varieties but genuine species, for cat and mouse preserve their specific peculiarities through generations, and never will one of their offspring suddenly be a leopard or a rat, nor will the offspring of a leopard or a rat ever become cat or mouse, although, hypothetically, a simple enlargement (or reduction in size) of their bodies and internal organs combined with some changes in colour, &c. would suffice to transform the one into the other. With regard to these animals, therefore, (and to almost all others as well) the size and some external features, like colour, &c. are not considered as determinative of varieties, but of species, and it is only the higher classificatory units such as genera, sub-families, &c. which become determined by the greater or lesser similarity in the internal organisation. Such principles of classification are, so far as I can see, universally followed by systematists with regard to the larger animals, and I do not see any reason, why they should not be applied to parasitic worms also; so much the more so, as careful observation and comparison reveal among them quite a similar gradual increase (or decrease) of structural differences as exists among the larger animals. I thus hold that it is simply logical and consistent to consider the two *Opisthorchis sinensis*-forms as two distinct species rather than as varieties of one species.

I have in the above explained the reasons for my opinion at some length, because the question what in helminthology is to be considered as a species still needs being definitely settled. It appears that quite a number of writers, zoological and medical, on helminthological subjects still cling to the old assumption that there exist comparatively few species of parasites only, but that these species are capable of varying in size and organisation to a considerable degree according to host, locality, &c. It is my firm conviction that this view must be altogether abandoned if helminthology is to become an exact science based on facts rather than on preconceptions inherited from our forefathers. Neither is it justifiable any longer to declare all at once two forms as identical simply because they agree in their general structure,

nor is it sufficient to-day to give of new forms of parasites a superficial account of some general characters, if such descriptions are to be of real scientific value. It would not be a difficult thing to enumerate dozens of modern papers of this sort; they look and read quite nicely, but nothing would have been lost had they never been published at all. I will not mention any names, nor is it my intention to blame or to offend those who do not share my scientific convictions, but I desire to convince them, in the interest of our particular branch of science, that by this time certain views considered as almost axioms by our predecessors can no longer be upheld.

The question as to the denomination of the two species of *Opisthorchis sinensis* in accordance with the modern rules of scientific nomenclature is easy to decide. There is no doubt that the large form is the same as originally found and described by McCONNELL; it must therefore retain the specific name *sinensis* COBBOLD. The smaller species is, unmistakeably again, the same as first recognised by BAELZ and distinguished under the designation *Distoma endemicum*; I therefore re-establish this specific name for it. Both species are members of a natural genus which might have been created before this on the ground of the peculiar configuration of the testicles, but becomes now more distinctly separated from *Opisthorchis* by the fact that two different forms exhibit the same character.

Clonorchis, n. g., *Opisthorchiidarum*.

Differs from *Opisthorchis* sensu strictiori by the fact that the testicles are not notched or lobate, but distinctly ramified, the branches, crossing the intestinal caeca on their ventral side and extending very near to the body margin. Another difference seems to lie in the shape of the excretory vesicle. In *Opisthorchis* s. str. this is Y shaped with a very long stem running in the form of an S through the space between the testicles and bifurcating at the posterior border of the seminal receptacle into two rather short branches from the tops of which the main excretory vessels arise. In *Clonorchis*, the Y shape of the vesicle is no longer clearly recognisable; the unpaired tube simply becomes somewhat widened at its anterior end assuming sometimes, but not always, the shape of an irregular triangle the two upper angles of which might be considered

as the homologues of the branches in *Opisthorchis* s. str. The main excretory vessels do not arise from these angles, but somewhat farther backward, at about the lower angle of the triangular widening of the tube (see fig. 4).

Type: *Clonorchis sinensis* (COBBOLD) 1875.

Clonorchis sinensis (COBBOLD) 1875.

(Synonym: *Distomum innocuum* BAELZ 1883.)

Full grown specimens vary in length from 13 to 19mm. and in breadth from 3 to 4 mm. At the commencement of the period of sexual activity the body has attained 12 to 13 mm. in length and 2·5 to 3 mm. in breadth; such specimens are recognisable by their uteri in which the ova are comparatively scarce in number and loosely grouped, i.e. with more or less wide empty spaces between the individual ova. The oral sucker averages 0·6 mm. in transverse diameter; the lowest figure I have found in one specimen (of about 60 measured) was 0·52 mm., the largest which is not uncommon 0·63 mm.; usual measurements between 0·58 and 0·62 mm. The ventral sucker averages 0·47 mm. in transverse diameter; lowest figure found in one case 0·39 mm., highest figure observed once also 0·52 mm.; usual measurements between 0·45 and 0·49 mm. The ratio of the suckers thus varies in the main between 15 : 12 and 16 : 12. The pigmentation, i.e. the deposit within the body parenchyma of numerous fine granules of a yellowish or brown substance, is very characteristic of this species. Sometimes it is so little developed that the preserved worms appear white and the pigment granules are only detected in cleared specimens with the microscope, yet may in other specimens assume such an extent that the parasites become perfectly opaque and of a rusty yellow, greyish, or brownish tint throughout. In cases of little pigmentation, the granules accumulate more especially behind the oral sucker in the sides of the pharynx and at the posterior extremity of the body; in a lesser degree along the intestinal caeca and round the outlines of testicles and seminal receptacle. The ramifications of the testicles are fairly long, in accordance with the greater breadth of the body; it may not unfrequently be recognised that in the anterior testicle the branches arise from four, in the posterior testicle from five main stems. All of them lie in about one plane which runs parallel to

the ventral surface of the body; there are however very often, in one testicle or in both, other branches or, more precisely, bulgings of one or two main stems which project towards the ventral surface and appear as thick, compact bodies with irregular outlines. They were already seen by MCCONNELL, but interpreted as the whole testicles, whereas in truth they are only parts of them, and are not even constant in their presence. The ovary is usually trilobate, but very often shows, in addition to the large lobes, three to six smaller ones which are sharply separated from the former. The yolk-glands vary somewhat in their extent. They normally reach from the level of the ovary to the ventral sucker, but it is not at all uncommon to see them, on one side or on both, commencing and ending somewhat farther forward or somewhat farther backward of the usual place. They present in *Cl. sinensis*, a striking peculiarity inasmuch as almost constantly a certain number of the groups of follicles of which they are composed remain undeveloped; sometimes one only on one side, sometimes more, on one side or on both; the bands represented by the glands as a whole showing thus one or several interruptions by empty spaces. In perfectly mature specimens the seminal vesicle extends about as far back as the middle of the uterus. The remaining organs do not show any peculiarities.

The average dimensions of the eggs are 0·029 mm. length and 0·016 mm. width; the limits of the former being 0·026 and 0·03 mm., and of the latter 0·015 and 0·017 mm.* In many specimens of the species the eggs show a distinct narrowing towards the anterior extremity, and their rather high lid is marked off by a sharply projecting brim. I have however also seen specimens in whose ova these peculiarities were but little pronounced (see fig. 7).

An interesting variation chiefly observable among the specimens of the Port Said case concerns the colour of the uterus as a whole. In some individuals, this is only a light yellowish-brown, even in the most anterior coils of the organ, whereas in other individuals the whole

* There exist, of course, among the immense number of ova in an individual worm always some which are either larger or smaller than the rest, or even evidently misshapen. In my opinion it is of no use to carefully record the measurements of these eggs also. For the description and definition of a species it is much more important to select for measurement those ova which appear to be normal and to present the size and shape typical for the species. It may be added in passing that young worms with few ova in their uterus usually do not afford normally-shaped and normally-sized ova.

uterus, with the exception of a few of the hindmost loops, appears almost black. As there is otherwise not the slightest difference between the examples of these two extremes--neither in the size of the body, nor in the size of the suckers, &c.--I do not quite know how to account for these variations. To a certain degree, they are evidently due to the number of ova contained in the coils, but to another degree just as evidently to a darker shade of the egg-shells themselves. Since the two extremes described above are not sharply separated, but connected with each other by numerous intermediary tints it is perhaps not unjustifiable to assume that the shell-substance elaborated by the shell-glands becomes darker with the growing age of the parasites, and it may be noted in this connection that the latter live in, and feed on, a coloured substance, the bile.

Clonorchis sinensis is chiefly a parasite of Chinese, but occurs rarely also in Japan--i.e. according to what we know at present. For it is not at all improbable that the worm will be oftener found as soon as attention is paid to its existence; it appears also not unlikely that it may be restricted to certain localities. Up to now it does not seem to have been found in animals.

Clonorchis endemicus (BAELZ) 1883.

This species practically takes the place of the true *Clon. sinensis* in the literature published after 1883 and is at present the one usually described in text books, &c. as "*Opisthorchis sinensis*." Its measurements are given by the authors as 10 to 13 mm. length and 2 to 3 mm. breadth. The smallest specimen actually at my disposal (it comes from the Annamite case described by BLANCHARD) is somewhat contracted and measures 6 by 1·8 mm.; my largest and fairly well extended specimen (of Japanese origin) measures 13 by 2·6 mm.; the remaining individuals range in size between these extremes. The oral sucker averages 0·43 to 0·45 mm. in transverse diameter; smallest diameter observed (in the smallest specimen mentioned above and another one (11 mm. long and 1·6 mm. broad) 0·37 mm.; largest diameter found (in a specimen 10·8 mm. long and 2·3 mm. broad) 0·5 mm. Ventral sucker averages 0·37 to 0·40 mm.; smallest diameters observed 0·33 mm. (in the smallest specimen) and 0·34 mm. (in another long 8 mm., broad 1·7 mm.); largest diameter observed 0·45 mm. (in the largest specimen above mentioned). The ratio of the

suckers varying between about 13 : 12 and 15 : 12 is therefore fairly similar to that in *Clon. sinensis*, but in actual size the suckers are quite different. Among the specimens at my disposal there is none with pigment-granules in its parenchyma. The yolk-glands normally have the same extent as in *Clon. sinensis*, but present similar variations in regard to the relative position of their anterior and posterior terminations as in that species. It may also happen that of the groups of follicles of which they are composed some remain undeveloped, but this occurs, so far as may be inferred from my limited material, much more seldom and the actual state of things is perhaps best described by saying that the absence of one or several groups of follicles is the rule in *Clon. sinensis*, and the exception in *Clon. endemicus*. The seminal vesicle ends (so far as visible) at about the limit between the first and second thirds of the length of the uterus; this character, however, had better not be considered as distinctive because it may vary.

The eggs have about the same length as those of the preceding species (0·026 mm.), but their width is decidedly somewhat smaller, amounting on an average to 0·015 mm. with the lower limit at 0·013, the upper at 0·016 mm. The narrowing towards the anterior end is in the main not so marked and the margin of the rather *flat* lid not so sharply projecting as in *Clon. sinensis* (see fig. 8); but these differences are on the whole very slight and not recognisable in every specimen. With regard to the colour of the uterus as a whole, variations may be found similar to those described in the preceding species.

Clonorchis endemicus, according to existing records, is very common in Japan, and presumably so in Annam and Tonkin too. In Japan the localities especially stricken with the parasite are situated on the sea-shore; it is possible therefore that its geographical range chiefly extends along the shore in the other localities also.

Apart from Man, the species is reported to have been found in certain animals of Japan, such as cats, dogs and hogs. Several specimens taken from the latter host, which I owe to the kindness of Professor JANSON, of Tokyo, are indeed *Clon. endemicus*, for they present all the peculiarities of this species. I am, however, somewhat doubtful with regard to the statement that it inhabits the liver of the cat also—at least as a normal parasite of this animal. In 1886, IJIMA after an examination of specimens derived from this source

writes that the worms " from the liver of cat showed slight differences inasmuch as they possessed very fine spines in the skin and were of smaller size than the former (viz. *Clon. endemicus* of the human liver). Perhaps this cat-distome from Okayama is to be considered as a distinct species, but I firmly believe that in Tokyo, *Dist. endemicum* does sometimes inhabit the liver of cats " (1886, p. 49). This latter possibility can certainly not be denied and is even most probable, but in cases of this sort *Clon. endemicus* appears to be merely an occasional parasite of the cat, whilst the smaller form mentioned by IJIMA probably represents one of its normal parasites. I have examined several cat-specimens which were sent from Japan to Professor LEUCKART in Leipzig years ago (by whom I do not know) and of which Professor CHUN kindly allowed me to take some in exchange for other parasites. They are unfortunately in such a poor state of preservation that not very much is left to be seen in them; the general impression they make upon the observer is certainly rather different from that of the genuine *Clon. endemicus*. If one considers, moreover, their small size (those in my possession do not exceed 4·5 mm. in length and 0·9 mm. in breadth, but are as it seems not yet quite full grown) the presence of fine spines in their skin as reported by IJIMA there is no doubt left that they cannot be *Clon. endemicus*.

A possibility worth being borne in mind in the examination of later cases of human Opisthorchiosis is that this Feline *Opisthorchis* (it cannot as yet be said whether it is an *Opisthorchis* or a *Clonorchis*) may occasionally occur in Man also, just as *Clon. endemicus* is occasionally picked up by cats and hogs. I am even inclined to think that certain statements in KATSURADA'S paper (1900, p. 481) alluding to three worms found in a young man of seventeen from the province of Saga, which presented an average length of 5·16 and an average breadth of 0·96 mm. only, may be explained by such occasional infection with the Feline species.

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EXPLANATION OF FIGURES

REFERENCE LETTERS.

App.—Lateral pouches of the oral sucker.
C.ex.—Lateral excretory vessels.
C.P.—Cirrus pouch.
D.ej.—Ejaculatory duct.
Disc.—Edge of the ventral disc.
Gr.—Retracted grooves ("pseudo-suckers").
Gr'.—The same organs, everted.
Int.—Intestine.
L.C.—LAURER'S canal.
l.Test.—Left testicle.
Met.—Metraterm.
Nerv.—Nervous system.
Oes.—Oesophagus.
Ov. Ovary.
P.ex.—Excretory pore.
P.Gen.—Genital aperture.
Phar.—Pharynx.
P.P.—Pars prostatica.
R.sem.—Seminal receptacle.
r.Test.—Right testicle.
Sh.Gl.—Shell gland.
Ut.—Uterus.
Vag.—Vagina.
V.ex.—Excretory vesicle.
V.ex.lat.—Lateral branches of the excretory vesicle.
Vit.—Yolk glands.
Vit.D.—Unpaired vitelloduct.
V.sem.—Seminal vesicle.

Fig. 1. *Eurytrema pancreaticum.* Specimen from Japan seen from the ventral aspect. Enlargement about 11.

Fig. 2. *Eurytrema coelomaticum.* Specimen from Hong-Kong seen from the ventral aspect. Enlargement about 11.

Fig. 3.—*Gastrodiscus secundus* seen from the ventral side. Enlargement about 11. The opening in the anterior part of the ventral surface leads into the genital atrium from the bottom of which the papilla bearing on its top the male and female genital openings arises.

Fig. 4. *Gastrodiscus secundus.* Median sagittal section; the organs cut in it have been completed from the neighbouring sections so as to give a general picture of the internal organisation. Enlargement about 16.

Fig. 5.—*Clonorchis sinensis.* Specimen from the Port Said case. Ventral aspect. Enlargement about 9.

Fig. 6.—*Clonorchis endemicus.* Specimen from Okayama, Japan. Ventral aspect. Enlargement exactly the same as that of the preceding figure so as to allow of an appropriate comparison. The unequal length of the intestinal caeca visible in this specimen is accidental and not present in other individuals.

Fig. 7.—*Clonorchis sinensis.* The chief forms presented by the eggs, each egg taken from a separate specimen of the worm. Enlargement ca. 990.

Fig. 8.—*Clonorchis endemicus.* Eggs figured in the same manner and under exactly the same enlargement as in the preceding drawing so as to allow of comparison.

PLATE VII.

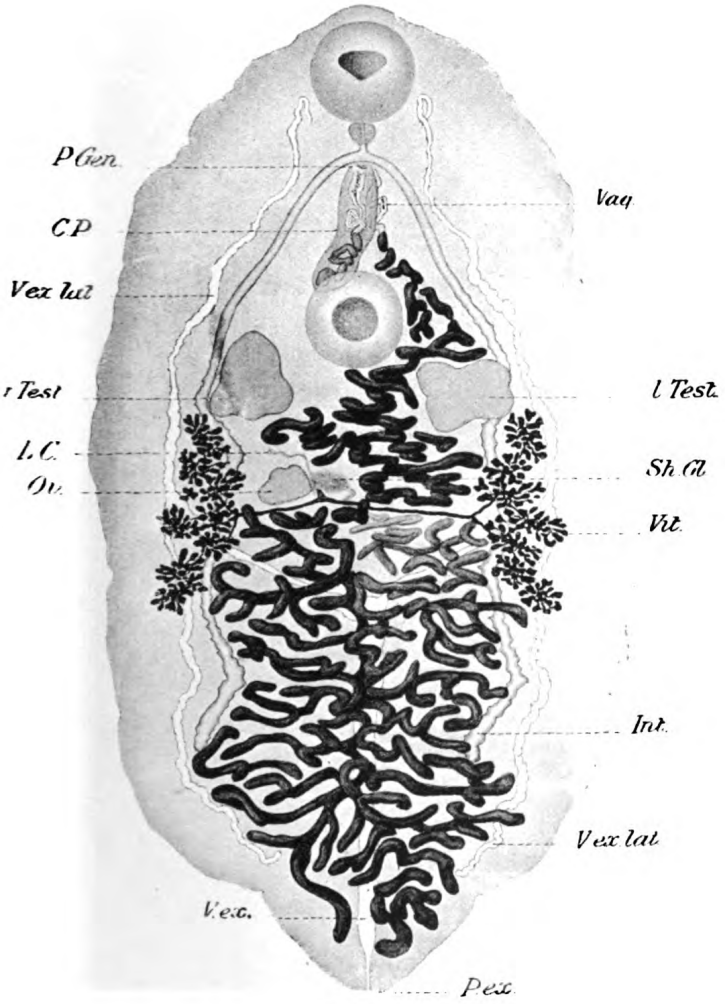


Fig 2.

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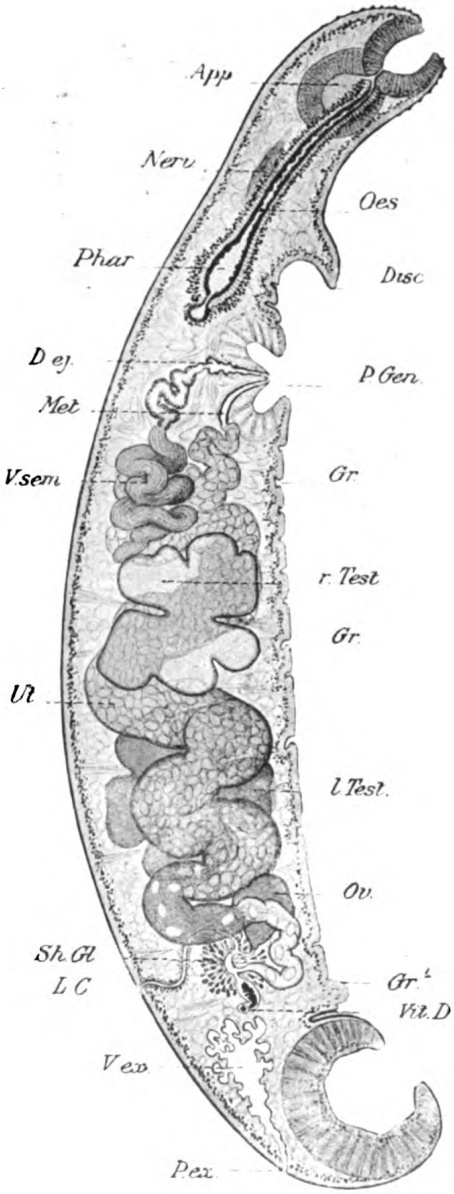

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Fig. 4.

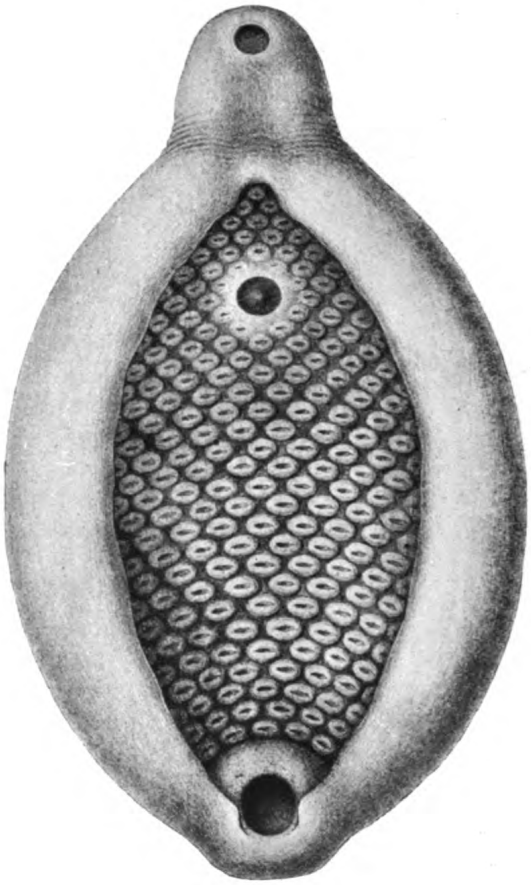


Fig. 3.

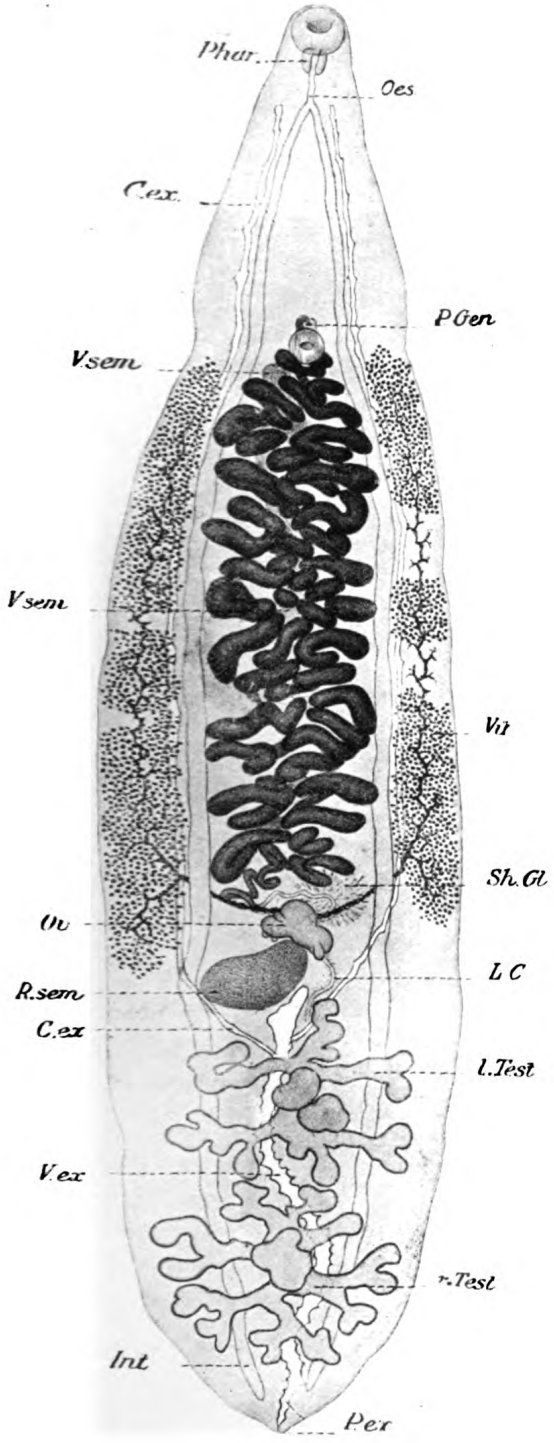


Fig. 5:

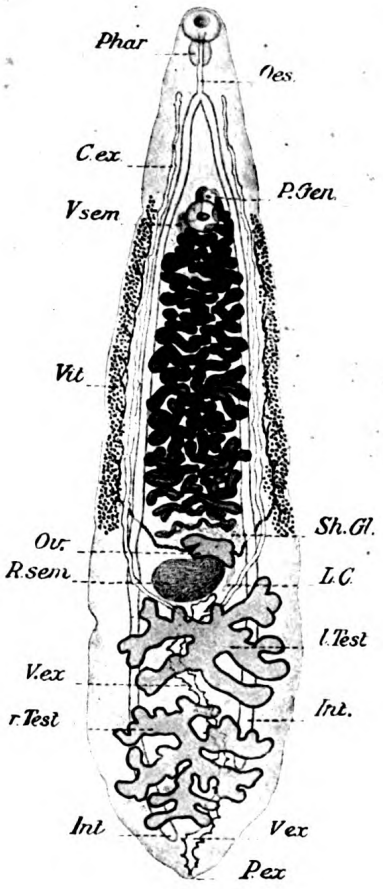


Fig. 6.

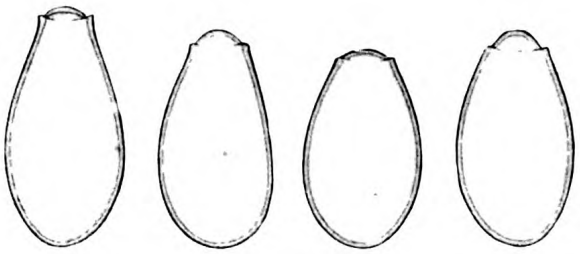


Fig. 7.

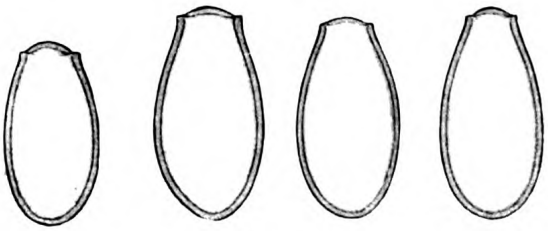


Fig. 8.

THE PRESENCE OF *SPIROCHAETA DUTTONI* IN THE OVA OF *ORNITHODOROS MOUBATA*

THE PRESENCE OF *SPIROCHAETA DUTTONI* IN THE OVA OF *ORNITHODOROS MOUBATA*

BY

CAPT. R. MARKHAM CARTER, I.M.S.

From the Runcorn Research Laboratories of the Liverpool School of Tropical Medicine

Koch, while discussing the question of the transmission of *Spirochaeta duttoni*, the organism of African relapsing fever[1] states that he finds on the surface of the ovaries of infected ticks large numbers of spirochaetes. He considers that these organisms multiply in this position in the tick. He further states that the eggs, when first laid, show scanty infection, whereas in those examined later he describes the occurrence of spirochaetes in vast numbers arranged in densely-tangled masses. This he considers an undoubted proof of the rapid multiplication of the spirochaetes in the eggs. Lastly, he again draws attention to the fact that ticks bred from batches of infected eggs transmit the disease.[2] The following observations were made with the object of confirming the second of these important statements.

Forty-two, naturally-infected, adult specimens of *Ornithodoros moubata*, collected in the Congo Free State, were fed on January 27th, 1906, upon an animal infected with *Spirochaeta duttoni*. These ticks were again fed on an uninfected rat on July 26th, 1906. This rat showed infection on August 1st, innumerable spirochaetes to the field were seen on August 11th. As no eggs were laid, the ticks were again fed; ovulation then commenced, but it was very scanty. Some twenty-six adults were therefore fed once more on an uninfected rat ten days later. Large numbers of eggs were now found in batches about the dish; 32 ova were taken from different collections of these eggs and microscopically examined to determine the presence or otherwise of the spirochaetes. Six of them were found to be infected.

The method used was as follows:—The egg, having been placed

in the centre of the slide, was pricked with a very fine capillary tube, and the contents were quickly withdrawn until the egg capsule became transparent and collapsed. The egg contents were then emptied, forming a tiny drop, on the slide, which drop was quickly smeared into a thin film with a fine-pointed, cold, glass rod. The more rapidly these stages succeeded each other, the greater the perfection of the difficultly made film. The preparation was quickly dried at 45° C. and was then fixed in absolute alcohol before being stained by a modification of Romanowsky's method;[3] finally the smear was examined with a Zeiss apochromatic 2 mm. objective and a No. 18 compensating eyepiece, fitted to a tube length of 250 mm. (× 2250).

The number of the spirochaetes found varied from but three in the whole contents of an egg, to as high as 45 counted parasites to a field, with, in addition, one or more spindle-shaped collections of innumerable organisms. As was observed by Koch it was found that most spirochaetes occurred in those eggs which had been laid longest.

Morphology of Parasites occurring in the Eggs (see plate X).

As well as the typical forms of the spirochaete, occurring as they did in large numbers, several peculiar varieties were also noted and will be discussed in detail below.

By far the majority of the spirochaetes were of the typical simple spirillar form. These varied in length from 5μ to about 20μ (single parasites), and were seen to occur singly and in groups of three to four. Occasionally, however, and this was seen in the most highly infected eggs, the spirochaetes were arranged in remarkable collections and were often parallel, spiral for spiral, with each other. These groups are occasionally spindle-shaped by reason of the decreasing length of those spirochaetes placed laterally as compared with the longest and usually more central parasites. Otherwise these grouped spirochaetes resemble an unbound bundle of sticks.

As a rule the typical[4] single spirochaete presents no evidence of a nucleus, blepharoplast or undulating membrane and no indications of lateral flagella. As a rule it seems to be a spiral, possibly flat and ribbon-shaped on section. One terminal point of the spirochaete is often deeply stained, but both or either may (fig. 7) stain faintly and taper to a point, possibly to form a terminal flagellum. In, especially, the long forms of the organism, transverse, faintly-staining sections

occasionally occur. These lightly-stained areas occur at about the middle of the parasite and occasionally excentrically in one or more places; this is also particularly noticeable in the exceptionally long forms. It seems as though the deeply-staining, central, chromatic core of the parasite were absent and the more lightly-staining periplast alone remains. The same appearance is seen at the extremities of some parasites; in many the condition is exaggerated until the whole chromatin of the spirochaete exists only as a fragmented succession of granules (figs. 8, 9, 24).

In figs. 19*a* and 19*b* are illustrated peculiar forms. They are large apparently single organisms, in which longitudinal binary division seems to be commencing. The undivided portion of these parasites is normal to within a short distance of its bifurcation. Here its chromatin becomes collected into one or two large granules. The divided portions lie side-by-side in parallel curves, and when fragmentation occurs in them it is noticeable that their corresponding granules lie directly opposite each other.

Swellings, usually occurring one to a parasite, are frequently seen in both normal and fragmented spirochaetes. They occur either terminally or in the length of the parasite and are of three types. The first is the smaller and consists of an oval, homogeneous, deeply chromatinic mass placed either centrally or laterally on the parasite (figs. 26 and 30). The second type is slightly larger, and consists of an oval, lightly-stained vesicle in which two chromatinic granules occur. It lies at the side of the parasite and is attached to it by a very fine connection (fig. 29). This form is by far the rarest; it was seen but once. The third and largest type presents the appearance of a vesicle containing a deeply-staining area, confluent with the length of the parasite, and a lightly-staining portion (fig. 29*a* and 29*b*).

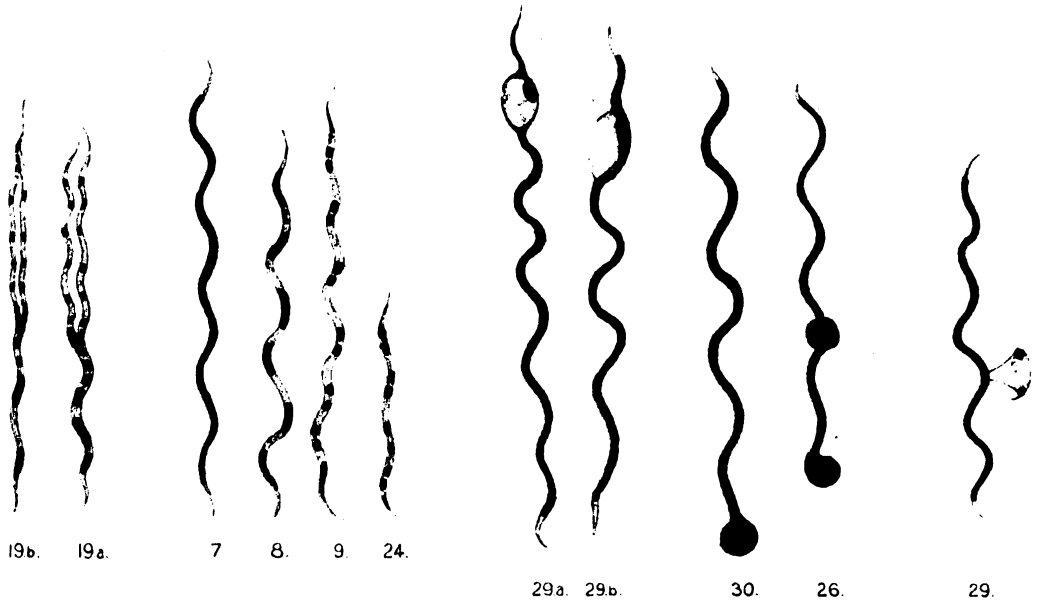
Spirochaetes presenting swellings of the first and last types frequently occurred in small groups. It is to be remembered that the observations recorded here were made on dried and stained specimens alone. I regret that lack of time prevents me from amplifying and confirming these suggestive observations by the examination of fresh specimens and of more carefully prepared material.

From these observations we conclude:—

1. That ticks infected by spirochaetes lay infected eggs.
2. That multiplication of the spirochaetes probably takes place in the eggs.
3. That morphological changes in the spirochaetes also occur in the eggs.

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X2500 about.

SPIROCHAETA DUTTONI IN EGGS OF ORNITHODORUS MOUBATA.

PLATE X.

A NOTE ON THE THERAPEUTICS OF TRYPANOSOMIASIS

BY

PROFESSOR BENJAMIN MOORE, M.A., D.Sc.

M. NIERENSTEIN, PH.D.

AND

JOHN L. TODD, B.A., M.D., C.M. McGILL

Thomas and Breinl* found that atoxyl would almost invariably drive the trypanosomes from the peripheral circulation of an infected animal, but, since the parasites frequently recurred, it seemed possible that they might exist somewhere else in the body of their host in a form uninfluenced by this drug. It was therefore determined to commence a series of experiments in which animals first treated by atoxyl were subsequently treated by some other drug. A variety of combinations have been tried. That which has been by far the most successful in the treatment of rats infected with *Trypanosoma brucei* is atoxyl, followed by small doses of bi-chloride of mercury after the trypanosomes have left the peripheral blood. Animal experiments are still in progress.

This communication is made in the hope of inducing those who have opportunities of treating patients to try the effect of atoxyl followed by bi-chloride of mercury on their patients.

* Thomas and Breinl; Memoir XVI of the Liverpool School of Tropical Medicine.

Volume I June, 1907 No. 2

ANNALS
OF
TROPICAL MEDICINE AND PARASITOLOGY

ISSUED BY

THE LIVERPOOL SCHOOL OF TROPICAL MEDICINE

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Regarding the Courses of Instruction proposed to be given by the Liverpool School of Tropical Medicine, and the Examinations for the Diploma of Tropical Medicine arranged to be held by the University of Liverpool during 1907 (subject to such alteration as may hereinafter be decided upon),

Lent Term begins January 15.
Lent Examination, March 25.
Summer Term begins May 1.
Summer Examination, July 15.
Autumn Term begins October 1.
Autumn Examination, December 16.

The full Course of Instruction is open to all qualified medical men, and the examination to all students who have taken out this full course.

Fee for the full Course of Instruction—Ten guineas.

Fee for the Examination—Five guineas.

Fee for the use of a School microscope during one term—Ten shillings.

For prospectus and further information, application should be made to the Dean of the Medical Faculty, University of Liverpool.

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1904	Augustine, Henry Joshua
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1904	Bruce, William James
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1906	Chisholm, James Alexander
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1904	Dee, Peter
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1906	Faichnie, Norman
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1904	Nicholson, James Edward
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EDITORIAL NOTICE

By order of the Committee of the Incorporated Liverpool School of Tropical Medicine, the series of the Reports of the School, which have been issued since 1899, will be followed, from January 1, 1907, by the Annals of Tropical Medicine and Parasitology, of which this is the second number.

The Annals are issued by the Committee of the School, and will contain all such matter as was formerly printed in the Reports—that is to say, accounts of the various expeditions of the School and of the scientific work done in its laboratories at the University of Liverpool and at Runcorn. Altogether twenty-one Memoirs, besides other works, have been published by the School since 1899, and of these ten, containing 519 quarto or octavo pages and 95 plates and figures, were published during the two years 1904 and 1905; and there is no reason to suppose that this rate of production by the workers of the School alone will diminish in the future. In addition, however, to School work, original articles from outside on any subject connected with Tropical Medicine or Hygiene may be published if found suitable (see notice on back of cover); so that, in all probability, not less than four numbers of the Annals will be issued annually. Each number will be brought out when material sufficient for it has been accumulated.

AN AUTOMATIC OILER FOR THE DESTRUCTION AND PREVENTION OF MOSQUITO LARVAE IN CESSPOOLS AND OTHER COLLECTIONS OF WATER

AN AUTOMATIC OILER FOR THE DESTRUCTION AND PREVENTION OF MOSQUITO LARVAE IN CESSPOOLS AND OTHER COLLECTIONS OF WATER

BY

E. H. ROSS, AND H. C. ROSS,

LATE SURGEONS R.N. AND MEDICAL OFFICERS' DEPARTMENT OF PUBLIC HEALTH IN EGYPT

(*Received January 16th, 1907*)

In the large towns of Egypt the breeding of mosquitoes is practically confined to the cesspools. With the exception of Alexandria there are at present no schemes for the disposal of sewage on modern lines, and therefore it passes into cesspools under or near the houses. These cesspools are practically the only collections of water where mosquitoes can breed, and it is to them that most attention must be paid in anti-mosquito campaigns.

Besides mosquitoes, all sorts of vermin live in the cesspools; and in plague-infected towns, the knowledge of the fact that rats have to depend on these collections of water for drinking purposes, is of the utmost importance.

In Port Said and Ismailia, where there have been successful campaigns, all cesspools are treated with a mixture of crude and refined petroleum once a week. This has up to now involved very considerable labour, a constant supervision, and a large expenditure of oil; and it was in the endeavour to overcome these difficulties that the following automatic oiler was designed.

Our aim was to produce an apparatus which would give a constant supply of oil to the surface of the water in the cesspools; which could be easily applied by unskilled labour (native); which could be cheaply made in any country; and lastly, one whose application would, if possible, reduce the cost of a campaign. We have found by experiment that this apparatus has fulfilled the requirements.

The apparatus is made of metal. We use the empty kerosene

tins in which the crude oil is supplied. The shape is that of a drum with two spouts, one at each end, and is shown in the drawing. It will be noticed that the spouts do not arise from the centre of the ends of the drum, but from one edge of them; are directly opposite to one another, and are numbered A and B. On the spout B a weight is suspended. All joints are soldered.

In A a sand filter is placed. This consists of a glass tube, with an internal diameter of one centimetre, containing sand. The tube passes through a cork which is pressed home in the spout. To make the sand filter:—Draw out some tubing to a point and cut off so that the lumen is about half a centimetre in diameter. Place a small plug of glass wool in the neck so formed and fill up two inches of the tube with fine sand. Then place another small plug of asbestos wool and again draw out the tube round it and cut off.

The whole apparatus should now be tested to see if it is tight. To complete it, a piece of galvanized iron wire is fastened to each spout, each piece being about two feet in length. The two pieces are joined together above, and from their junction a wire passes upwards which must be long enough to reach from nearly the bottom of the cesspool to the manhole door. This appliance when filled with water will immediately sink, assuming a horizontal position under water; but when filled with oil, would float were it not for the weight on the spout B. Consequently, when filled with oil and dropped into water, it will sink to the bottom, but the end A will be pointing uppermost, and the oil will pass out through the sand filter at the rate of about 50 cubic centimetres a day. The oilers hold nearly $3\frac{1}{2}$ litres of oil.

To sink an oiler in a cesspool the following procedure is adopted. A nail is driven into the wall of the manhole just under the lid. The oiler is held with the spout B upwards and oil is poured into it by means of a funnel. When full it is allowed to rest horizontally on the ground with the spouts on top so that little or no oil is spilled. (Fig. 1.)

The apparatus is raised by the wire and lowered gently into the cesspool until it touches the water. It remains horizontal until the water is reached, when it immediately assumes its vertical position as it sinks under the surface. (Fig. 2.) No oil will pass out until the air-bubbles coming out of the end A have ceased, when the exudation of oil begins.

The wire is payed out until the weight is felt to rest on the bottom of the cesspool; it is then pulled up for about one foot and fastened to the nail. In this way the oiler will rest about eighteen inches from the bottom of the cesspool. (Fig. 3.) Two litres of oil are then thrown on to the surface of the water to kill the existing larvae, and the manhole door is closed. In the oil thrown on to the surface it has been found better to mix five per cent. of powdered resin, as this materially prevents evaporation. But resin must not be mixed with the oil inside the oiler, as it may clog the sand filter.

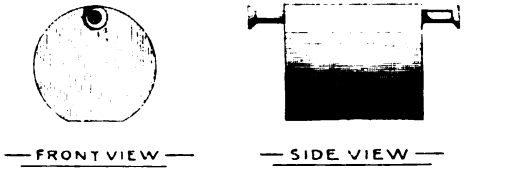
The exudation of oil goes on at the rate of about 50 cubic centimetres a day for nearly two months. As oil comes out of spout A water enters the oiler through the spout B, and therefore it becomes heavier daily and consequently assumes a more horizontal position, until in about two months, when there is about three-quarters of a litre of oil left, the oil comes out of the spout B and the oiler is suddenly emptied of the mixture. (Fig. 4.) So that in the eighth week of the oiler's existence in the cesspool there is a sudden and large ejection of oil on to the surface of the water.

The reason we have designed this is that if the cesspool is a large one and the exudation of oil hardly sufficient, any larvae which may have evaded the oil towards the end of the two months will be destroyed.

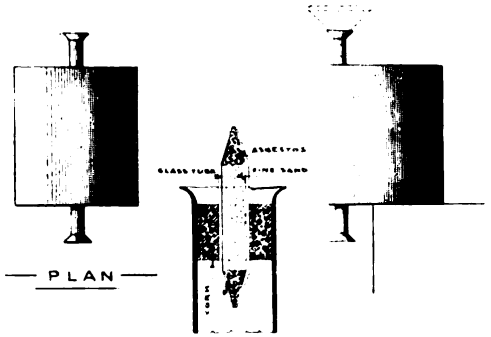
In ten weeks the oiler is removed and replaced by a new one. In Egypt the cost of each oiler complete without oil is sixpence. The apparatus has been given a fair trial, and has been found to be successful. It has saved a large amount of labour and oil, and will shortly be used extensively in Cairo and its suburbs.

AUTOMATIC OILER

FOR CESSPOOLS



FRONT VIEW — SIDE VIEW



PLAN

SECTION OF GLASS FILTER

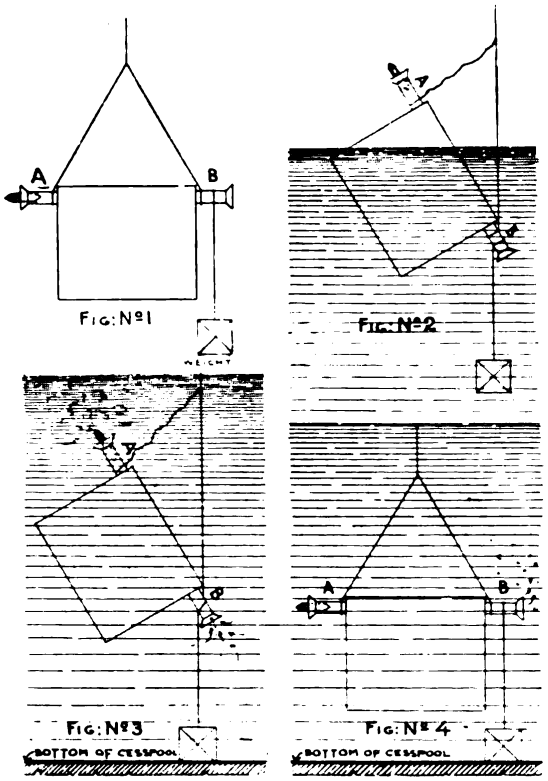


PLATE XI

THE ANATOMY OF THE PROBOSCIS OF BITING FLIES

THE ANATOMY OF THE PROBOSCIS OF BITING FLIES

BY

J. W. W. STEPHENS, M.D. CANTAB.

AND

R. NEWSTEAD, A.L.S., F.E.S.

(*Received May 11th, 1907*)

PART II.* STOMOXYS (STABLE FLIES)

As far as we are aware, there have been only two descriptions, in recent years at least, of the anatomy of the mouth parts of *Stomoxys*. The first, by Hansen,[1] is, as far as it goes, a good and accurate account, but it is incomplete in many respects, even in regard to important structures, which we should have thought it impossible to overlook. Moreover, sections were not cut by this author, and without these, it is, in our opinion, impossible to understand the relationship of the parts. The second description is by Giles.[2]. We feel bound to point out that this account is misleading and inaccurate; and that the same criticism is equally applicable to his description of *Glossina* (*l. c.*)

METHODS.—For section cutting we find that a combined celloidin-paraffin method gives the best result. In this way it is possible to get intact sections of the chitin, otherwise a difficult matter. Owing to the opacity of the proboscis, examination with high powers, of specimens mounted *in toto*, is almost impossible; but the whole proboscis can be rapidly rendered transparent to any desired extent by bleaching in chlorine gas: the chitin not being at all affected.

* Part I, *Glossina* (Tsetse-flies), was published in Memoir XVIII, 1906, Liverpool School of Tropical Medicine.

The material at our disposal having been fixed in alcohol has not been in a sufficiently good condition for an accurate study of the soft cellular structures of the labella, e.g., the nerves. We hope eventually to examine these structures in detail.

The species examined by us has been *Stomoxys calcitrans*, Linn. (the common "stable-fly"), caught in England.

The proboscis of *Stomoxys* (fig. 1), as in *Glossina*, consists of three parts: the lower lip or labium (*c*), the hypopharynx (*b*), and the upper lip or labrum-epipharynx (*a*). The actual length from the base of the bulb to the tip of the labella measures 2·1 mm.; the dimensions just behind the fork are 130 μ vertically and 150 μ horizontally.

THE LABIUM (fig. 1, *c*) is relatively stout and thick, with the basal portion swollen and gradually tapering towards both the proximal and distal extremities, but less so basally. Externally in its entire length, as far as the labella, the labium is transversely and regularly striated or latticed, so that the edges of preparations mounted entire appear faintly crenulated. Scattered over the labium are also a number of short fine hairs, set on transparent spherical areas; more numerous, however, on either side of the dorsal groove. Immediately behind the fork of the labella (fig. 5, *e*), and articulating with it, are two conspicuous ventral sclerities (fig. 5, *f*).

LABELLA (General account). While the arrangements of the parts in the labium is easily understood, that of the labella presents, on the contrary, considerable difficulty, and we must consider the structures in detail. The labella are bilaterally symmetrical. They consist of an external and internal wall on either side (fig. 20), separated by soft tissues. The two halves are at first separate, near the base the sides join ventrally (figs. 21, 22), but remain open dorsally. When the labella are in the resting position, the internal walls consist of two strong chitinous plates (fig. 20), which we may divide into a dorsal and ventral area. The dorsal half (*k* in all the figures) consists of a flat plate, which is the true plate of the labella, while the ventral half is highly specialized to form a series of teeth (*j* in all the figures) and other structures, which we shall consider in detail later. If we place our two hands back to back, the fingers will represent the ventral half (the teeth, etc.), while the palms back to back represent the dorsal half. Further in the process of eversion the ventral half is capable

of being bent outwards and backwards. If we now flex the fingers this gives approximately the position that the ventral half of the plate (the teeth) takes when the proboscis is everted.

The two bilateral inner walls are at first approximately parallel. If we now trace them backwards, we find that at first the inner walls approach one another ventrally so that they now enclose a somewhat V-shaped space (fig. 22). The dorsal half of the plate (*k*) grows downwards at the expense of the ventral, so that in fig. 21 we have only a few remnants of the ventral half left (and here the two sides are united ventrally), and eventually these disappear, so that we have on either side what was the dorsal half now occupying the whole inner side (fig. 22, *k*). These two sides now become fused ventrally by a median structure (figs. 23, 24), the axial apophysis (*q*), which serves to bind them together.

Dorsally the inner sides now become replaced by new structures appearing, viz., by two lateral chitinous rod-like sclerites, seen in cross-section in figs. 21-24, *r*, and medially a structure (r^1) appears which will form eventually the floor of the labial gutter when these two rods and the median piece fuse to form a single trough.

LABIAL GUTTER (*r*, r^1 in all the figs.). The commencement of the labium can be seen in a cleared proboscis as a transverse line stretching across (above) the middle of the axial apophysis. In sections it appears at about the level of the tips of the fork as a slightly hollowed-out free chitinous piece. About the same time appear, dorsal to it, two chitinous rods (*r*) seen in cross section in figs. 21-28. These rods are hooked at their edges, and to these edges are attached ligamentous tissues continuous with the chitin externally. At first there is only a fibrous junction with the ventral piece, which soon expands laterally and dorsally, and it is not till some way down that the fusion takes place and the canal assumes its complete shape. In the region of the base of the fork the canal has developed a ventral projection (fig. 28), and this goes on developing until the appearance of a row-lock is obtained, as seen in fig. 29, which again disappears in the bulb.

The axial apophysis expands posteriorly into a rounded mass seen in fig. 25, and by this level the original lateral internal walls of the labella have completely disappeared, the hind portion of the apophysis (*q*) now lying free, imbedded in soft tissues.

THE AXIAL APOPHYSIS (*q* in all the figs.) is a somewhat pear-shaped median structure, the tip of which lies completely free between the two inner walls (fig. 22); its middle portion is, as we have just said, fused on either side to the inner wall. Posteriorly again it expands into a bilobed bulbous base which instead of being free is surrounded by soft tissues. It is, in fact, suspended by fibrous tissue to the under surface of the dorsal gutter (figs. 23-25). It often shews evidence of its bilateral origin in the existence of a median cleft (fig. 24) potential or actual.

THE FORK (*e* in all the figs). When we have reached the free end of the axial apophysis there now appears on either side of it the tips of the fork cut in transverse section and attached to the tips, we see also the tendons (*s* in all the figs.) which pull on these tips (fig. 22). As we proceed basally we find these chitinous arms of the fork approaching one another ventrally (fig. 25), finally fusing, as shewn in fig. 26 (also fig. 12, eversion). The base of this fork rests on two ventral sclerites (fig. 6, *f*), and in transverse section still nearer the base the only internal chitinous structures are those forming the dorsal canal and the ventral sclerites. In this section (fig. 28) we see stretching from the canal to the ventral sclerites powerful muscles (s^1), and laterally we see the tendons (*s*) cut across. We may note here, also, the incurving and thin character of the chitin of the lateral wall at this point. The ventral sclerites now decrease in size, the muscles are replaced by various other groups of muscles and the dorsal canal has practically assumed the shape it has for a great part of the rest of its course.

By this time also the epipharynx has appeared, closing in the canal dorsally (fig 28, *a*). We may now consider some of these parts more in detail.

THE EPIDERMIS forming the external walls of the labella (figs. 2, 4, 5) presents a finely reticulated surface due to small polygonal squamae, which structure admits of considerable folding of the integument during the eversion of the labella (figs. 10-14). When the labella are closed, two deep oblique, bilateral folds are seen dorsally (fig. 4); the first (c^4) commencing near the apex and terminating at or near the tips of the fork; the second (c^5) commencing behind the lateral group of hairs (*vide infra*). Ventrally there are no

folds in front of the fork, but there is a deep oblique fold immediately behind it (fig. 5, c^7). When the labella are everted (figs. 2, 3, 10, 11, 12, 14) the folds become very complicated and difficult of interpretation, but there are three deep ones in front of the pulley. Dorsally, a little posterior to the point where the tip of the fork impinges against the inner surface of the external wall (fig. 14, *e*), a complete pocket or *cul-de-sac* is formed (fig. 13). Ventrally there is also another deep fold which, when seen in transverse section (fig. 12, c^7) appears as a projecting and slightly divergent arm.

EXTERNAL HAIRS. These occupy three principal areas:

(1) A ventral series (d in all the figs.) immediately within the thin flexible terminal flanges of the labella (figs. 2, 3, 5, 6, 9, &c.). There are from 15 to 16 of these on either side, three of which are much longer than the rest; all are stout, curved, rest on broad cylindrical bases, and are equidistant, with the exception of two or three, which give the otherwise complete line a somewhat irregular appearance. These hairs are easily traced in all preparations; and during eversion they are directed backwards instead of forwards, forming an almost complete fringe below the teeth. Lying folded back on the proboscis the tips of the largest reach nearly as far backwards as the fork.

(2) A dorsal bilateral row of five or six setae (d^1 in all the figs.). These are situated partly within the apical fold of the labella (fig. 4), and when the labella are everted they take a more or less transverse position, with the tips of either row overlapping or meeting along the median line of the groove of the labella, forming a ladder-like arrangement (figs. 2, 14). These hairs can be seen only in preparations that have been thoroughly bleached.

(3) A lateral group of minute hairs (d^2 in all the figs.). These lie immediately behind the tip of the fork when the labella are everted (figs. 2, 14), occupying a sub-dorsal position; but when the labella are closed, this group takes a more lateral position (figs. 4, 6) in the region opposite the sclerites which form the walls of the dorsal canal of the labium. The individual hairs are extremely minute (the length less than the width of the clear area in which they are placed) and difficult to see; each one is placed in the centre of a large, clear circular or "gland-like" area, which is well marked off from the surrounding chitin, and thereby rendered conspicuous.

VENTRAL FLANGES OR LIPS OF THE LABELLA (c^2 in all the figs.). These are extremely thin and flexible but are strengthened by several rows of strongly chitinised scales, of which the external row is much the larger. When the labella are closed these flanges or lips meet together and completely enclose the internal mouth-parts (fig. 19); but when eversion takes place they occupy a dorso-lateral position, and the edges are then projected upwards and dorsalwards (figs. 2, 3, 10, 11), resting practically on the sub-lying chitin of the external wall.

THE FORK (e in all the figs.). A proboscis mounted in the dorso-ventral position, as in fig. 5, shows at the base of the cleft a solid chitinous median structure, the axial apophysis (q), and some way behind this a U-shaped piece of chitin, the fork, with its concavity opening forward. With this, are seen articulating two lateral sclerites (figs. 5, 6, e^1) and posteriorly two ventral sclerites (figs. 5, 6, f). The joint between the base of the fork and these ventral sclerites is fringed with rows of hairs (fig. 6, f^1).

Apically the fork, which is, as we shall see, an internal structure, is prolonged as two flanges (fig. 5), which are continuous with the external chitin. Laterally the fork articulates with the two lateral sclerites, as shown in section in fig. 25, e^1. These lateral sclerites are directly continuous (fused) with the chitinous integument, so that by this connection, and by means of the direct continuity at its tips, the fork is enabled to produce eversion of the labella when acted upon by the powerful muscles of the bulb. Stretching across between the arms of the fork is a band of fibrous tissue (fig. 25). This is first seen when the hind end of the axial apophysis disappears. This band serves to strengthen the arms of the fork, and by its attachments to the fibrous tissue in which the peg lies must exercise traction on the peg itself when the fork takes the vertical position. In the everted position of the labella we see that we lose this appearance of a fork, and this organ is seen stretching across almost as a band (fig. 3,e). This change of position, we shall see, is produced by the tips moving dorsally and posteriorly (backwards), so that now in a transverse section the fork is cut along its length (fig. 12) instead of being cut transversely (figs. 21, 22). In cleared specimens also the tendons can be seen (fig. 6s), which are attached to the fork at its tips (fig. 3, s).

INTERNAL ORGANS OF THE LABELLA

These consist of seven different organs and are bilaterally symmetrical (fig. 8).

(1) THE TEETH (*j* in all the figs.). There are five of these and a small accessory or rudimentary one. They are relatively very large, the length of an individual tooth being equal to about one-fourth the entire depth of the labellum. They are articulated to an extremely flexible, narrow plate, which is in turn articulated to the true plate of the labellum. The first (proximal) and second tooth have each a strong lateral process (tooth) on the lateral proximal margin, and the tips of each are serrated on both sides; the three succeeding teeth are similar in structure, but the lateral deep dentitions are now on the opposite sides. In the everted position the teeth are freely exposed, widely divergent, and project at right angles and even backwards and dorsalwards. It is evident from their position and extent of motion that they act as the chief and, in fact, the only cutting organs of the proboscis.

(2) THE PETIOLATED BLADES (*l* in all the figs.). These are long, curved, blade-like organs covered with minute hairs on the ventral concave surface. All are attached to long and extremely slender stalks arising from strongly chitinised projections of the narrow tooth plate. They are grouped in the following order:—At the proximal extremity, exterior to the tooth, are two groups; the first consists of two or three very small blades attached to a relatively strong branched stalk; the second group consists of seven or eight blades, some of which are scarcely larger than those of the first set; the main stem in also stout, but the lateral stalks carrying the blades are extremely slender. Between each tooth is a set of four blades arranged in two pairs, the blades of each pair being widely separated, so that they resemble, somewhat, the arms of a lyre; the blades partly overlap, but the tips of the lower pair extend considerably beyond the upper pair, and when the labella are closed they reach as far as the basal row of external squamae forming the extreme edge of the labella (fig. 10, in dotted lines). Anterior to the last (fifth) tooth are three more sets of blades; the first set (between the fifth and the accessory tooth) consists of six or seven blades; the second of a similar number, and the third of four or five, of which three resemble

those carried on the first proximal stalk. The stalk to which the blades are attached, more especially those between the teeth, are, as we have already stated, extremely slender. They are, in fact, scarcely thicker than the fine marginal hairs of the labella, are filamentous in character, and in stained preparations present a well-marked granular appearance. The relative position of these organs in the everted sections may be seen in figs. 2, 3, 10, 11.

(3) ROD-LIKE HAIRS (*m* in all the figs.). These are dark in colour, in fact almost black; they are long, more or less curved, bluntly rounded at the tips, and apparently grooved on one side. They are articulated to strong cylindrical bases similar to those of the ventral external hairs, are fixed in a semi-erect position, and are arranged in the following order:—One opposite the tip of the first (proximal) tooth, a pair opposite each of the second, third, fourth and fifth tooth, and one opposite the accessory tooth, making ten in all. As these organs are inserted on nucleated cells they are probably sensory in function. The position of these organs, when the labella are everted, is shown in figs. 2, 3, 10, 11.

(4) THE OPERCULATE HAIRS (*n* in all the figs.). These are minute cylindrical organs with suddenly truncate extremities, from the edge of which there arises a flat oval-shaped lid or flap, which stands out an angle of 45° or more from the margin. There are five of these curious organs, one opposite the tip of each of the five large teeth; they form a regular curved line and are fixed to the integument about mid-way between the ends of the teeth and the tips of the petiolated blades. The position of these organs in the everted labella is shown in fig. 11.

(5) BIFURCATED HAIRS (*o* in all the figs.). These are rather long, swollen basally, and divided a little beyond the middle, the branched portions being extremely slender. They form six well-marked and almost continuous groups, slightly in advance of the operculate hairs, each group having a tendency to arrange itself into rows (fig. 8). They occupy a lateral position in the everted sections (figs. 10, 11), just below the ventral flange of the labella.

(6) THE ANTERIOR DORSAL FRINGE OF HAIRS (*p* in all the figs.). These hairs clothe the whole of the extreme anterior dorsal margin of the labellum (fig. 8). This fringe is broad basally, but gradually

narrows as it approaches the ventral margin. In everted sections (figs. 10, 11) these hairs project dorsally into a pseudo-cavity formed by the over-arching wall of the epidermis; and when thus in apposition they shut off the cavity of the labial trough.

(7) PROXIMAL FRINGE OF HAIRS (p^1 in all the figs.). These are similar to the former, but occupy a relatively small space on the proximal margin.

LABRUM (figs. 14-19)

This commences immediately behind the fork. Dorsally and laterally for its whole length it is covered with spinose hairs directed apically. At the apex itself (fig. 16) the hairs are replaced by scales. Near the tip, the labrum has ventrally a ridge or chitinous band extending across it (fig. 19), on which are situated four hairs evidently sensory in function, as nerve endings can be traced into them. Behind this point the sides widen slightly and along the interior are here arranged in rows, a series of hairs (a^2) on tubercles.

The labrum is at first concavo-convex in section, the convexity near the tip forming a kind of ridge (fig. 17). A little further down it has the appearance of an **U** fitted into a **V** (fig. 18). In the cavity between the two there appear early soft structures which later become developed into well-marked muscular bands connecting the lateral walls of the labrum (figs. 29, 30, s^4). The sides of the labrum become soon expanded into lateral flaps, which serve the purpose of keeping the hypopharynx in its place, the flaps fitting on to the expanded sides of the latter (figs. 29, 30). Eventually the ventral wall of the labrum becomes closed ventrally, forming now a chitinous tube, while the dorsal wall becomes fused and lost in the substance of the bulb (fig. 31).

THE HYPOPHARYNX (b in all the figs.)

This organ commences at about the same point as the labrum. It is a much smaller and weaker tube. At the extreme tip it appears to be membranous in character and to retain no very definite constant shape in preserved material. (We would suggest that this flaccid condition of the extreme tip is to prevent ingress of fluid, e.g., blood during feeding, while permitting freely of egress of saliva.) It

occupies the dorsal gutter of the labium, and is kept in this position by the pressure of the lateral arms of the labrum which stretch over on either side for a short distance (figs. 29, 30).

In section near the tip it appears as a chitinous hoop, forming a circle complete for about seven-eighths of its circumference. At first the hoop has near the apex on each side a little tubercle, and stretching across between these tubercles is a thinner chitinous membrane completing the tube dorsally. As we get nearer the base there is a distinct separation in the walls of the chitinous ring in the position where the tubercles first appear, so that a cavity now exists in the sides (fig. 29). This goes on increasing till eventually we have the chitinous circular inner tube lying within a sausage-shaped outer covering, forming a projecting angle on either side (fig. 30, *b*).

At its base the ventral wall of the hypopharynx becomes fused with the bottom of the labial gutter. The beginning of this is shown in fig. 30. This proceeds until the process is complete on either side. The fused wall is then absorbed, and thus permits of the inner (hypopharyngeal) tube escaping through ventrally. At the same time the dorsal wall of the hypopharynx becomes fused with the ventral half of the epipharynx, forming now the pharyngeal tube (fig. 31), while the dorsal half of the epipharynx fuses laterally with the sides of the labium, so that the whole of the epipharynx and hypopharynx, with the exception of the dorsal wall of the former, have now become internal structures.

MUSCULAR SYSTEM (s, s^1-s^5 in all the figs.)

In dissecting the proboscis it is often possible to separate out two large separate masses of muscle which terminate in tendons (s). These latter are attached to the tip of the pulley and are seen in all the sections behind this plane until we reach the bulbous portion of the proboscis. Here the tendons give place to the muscles from which they arise, which consist of two masses filling posteriorly practically the whole of the ventral portion of the bulb. These muscles we may describe as the longitudinal muscles. Besides, we have on either side of the median limb of the labial gutter a group of oblique muscles (s^3) forming a broad band, arising from the chitinous integument and inserted into the median limb (fig. 29). Dorsal to this group, and separated from it by the tendon, there is a second oblique group (s^2)

converging from the outer wall to the base of the gutter. Both these groups can be traced as far forwards as about the base of the ventral sclerites. Anterior to this point we have a well-developed band of dorso-lateral muscles stretching from the canal dorsally to the ventral sclerites ventrally (fig. 28, s^1).

MUSCLES OF THE LABRUM. We have also sub-median muscles present in the cavity of the labrum stretching from the outer to the inner walls (figs. 29, 30, s^4).

INTERNAL LAMINAE (fig. 28, s^5). In sections in the labella region, where the dorsal fold is well marked, there appear beneath the fold of the integument peculiar yellowish or brownish granular bands, of irregular appearance following the outline of the integument. They often show little irregular lateral projections. They represent transverse sections of plates. They are seen in sections as far as the ventral sclerites. They are, moreover, very well developed also in the cavity of the labrum in its basal portion. We are unable to suggest what may be the function of these bodies.

MECHANISM OF THE PROBOSCIS

With regard to the mode of action of these muscles, we consider that the function of the longitudinal group is to erect the fork and, by so doing, to produce by the traction on the external chitinous wall, eversion of the labella and a resulting spread-out condition of the teeth.

The oblique muscles' function is, we believe, to produce a semi-rotatory movement of the proboscis[3] (and labella), so that the teeth are enabled to exert a cutting action on the skin, the motion being a to and fro one.

The cutting weapon of the proboscis is undoubtedly the teeth. These are capable of a variable extent of eversion, and while they are in action the other structures anterior to them, petiolated blades, rod-like hairs, etc., are folded back in a plane below the actual tips of the teeth, and so escape damage.

The tip of the proboscis then can only cut in the everted position. We have in this condition of everted teeth an admirable weapon for boring a hole into the skin. The arrangement of a carpenter's augur with its cutting flanges on either side is almost an exact parallel, though the motion here is a continuous rotatory one, and not a to and fro

one as in the case of the fly. A study of the motion of proboscides in decapitated flies shows what an extremely flexible and mobile organ the tip of the proboscis (labella) is. It is capable of extensive rotation, so much so that the dorsal and ventral positions can almost be reversed. There is, of course, just as in *Glossina*, no motion possible of individual parts along the long axis, owing to the fusion of all parts at the base. A close examination of the labium during life will, however, show that its sides are to some extent flexible, being pulled on by the various muscles attached along its length.

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EXPLANATION OF REFERENCE LETTERS

External Organs.

- a Labrum-epipharynx.
- a^1 Ventral ridge of labrum bearing sensory hairs.
- a^2 Lateral ventral hairs to labrum.
- a^3 Apical scales to labrum.
- a^4 Dorsal hairs to labrum.
- b Hypopharynx.
- c Labium.
- c^1 Labella.
- c^2 Ventral flange or lip of labella.
- c^3 Epidermis of labella.
- c^4 First dorsal groove in epidermis of labella.
- c^5 Second dorsal groove in epidermis of labella.
- c^6 Third dorsal groove in epidermis of labella (produced in eversion).
- c^7 Ventral groove in epidermis of labella.
- c^8 Fold forming cul-de-sac.
- c^9 Fold behind fork.
- d Ventral external hairs.
- d^1 Dorsal external hairs.
- d^2 Lateral group of external hairs.
- e Fork.
- e^1 Lateral sclerite to fork.
- f Ventral sclerites.
- f^1 Hairs at apex of ventral sclerites.
- g Palpi.
- h Apodeme of labrum.

Internal Organs.

- i Tracheal sac.
- i^1 Tracheal tube.
- j The teeth.
- j^1 Accessory tooth.
- k Tooth plate.
- l Petiolated blades.
- m Rod-like hairs.
- n Operculate hairs.
- o Bifurcated hairs.
- p Anterior fringe of hairs.
- p^1 Proximal fringe of hairs.
- q The axial apophysis.
- r Dorsal hooked sclerites forming upper portion of the labial gutter.
- r^1 Sclerite forming base of labial gutter.
- s Tendon.
- s^1 Dorso-ventral muscles.
- s^2 Median oblique muscles.
- s^3 Ventral oblique muscles.
- s^4 Transverse muscles in cavity of labrum.
- s^5 Internal lamina.
- t Chitin cells.
- u Interlocking spines.

EXPLANATION OF PLATE XII

Fig. 1.—External mouth parts of *Stomoxys calcitrans* seen from the side, × 36, about. *a*, labrum; a^1, ventral ridge of labrum bearing sensory hairs; a^4, dorsal hairs; *b*, hypopharynx; *c*, labium; c^1, labella; *g*, palpi; *h*, apodeme of labrum; *i*, large tracheal tube or sac.

Fig. 2.—Labella everted, dorsal aspect. The left half shows the characteristic scale-like structure of the epidermis. × 200.

Fig. 3.—Labella everted, ventral aspect. Details of epidermis omitted. × 200.

Fig. 4.—Labella closed, dorsal aspect. × 200.

Fig. 5.—Labella closed, ventral aspect. × 200.

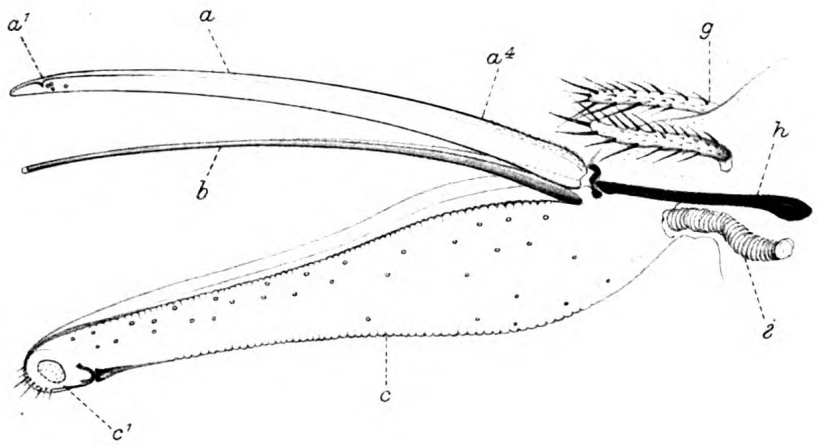


Fig.1.

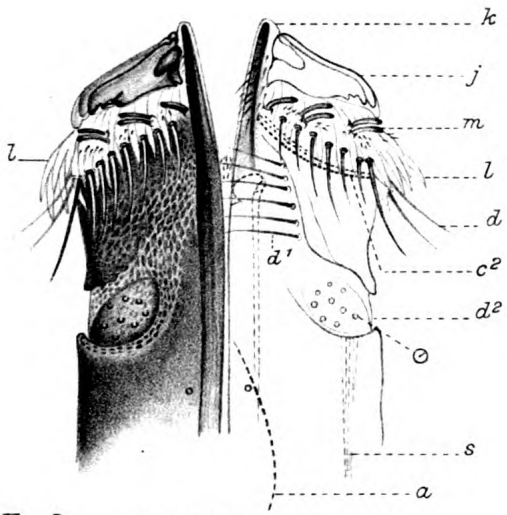
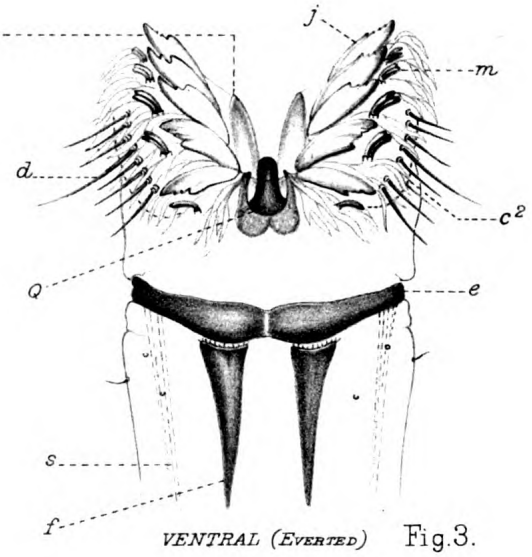


Fig.2. DORSAL (EVERTED)



VENTRAL (EVERTED) Fig.3.

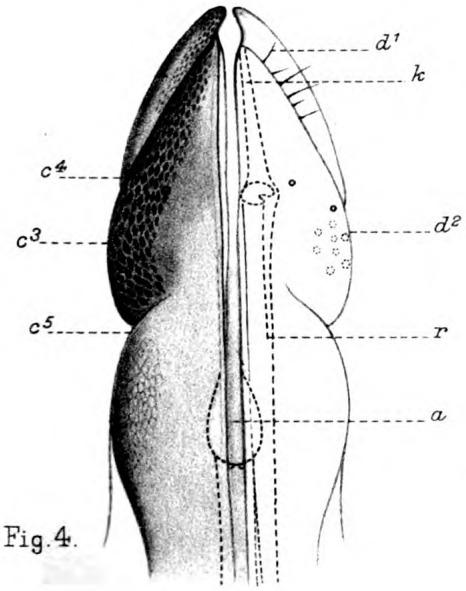


Fig.4.

DORSAL (CLOSED)

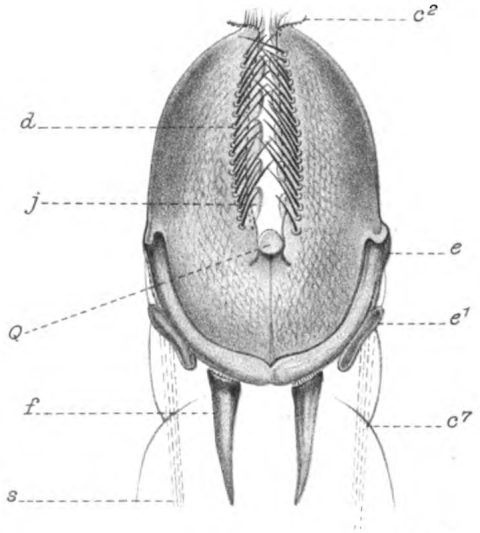


Fig.5.

VENTRAL (CLOSED)

R. Newstead ad nat. del.

West, Newman lith.

EXPLANATION OF PLATE XIII

Fig. 6.—Plan of labella (closed) as seen from the side, showing the relative position of the internal and external organs. × 200.

Fig. 7.—Internal organs of right labellum and distal portion of labial trough (r, r^1) as seen in optical section after dissection. × 250, about.

Fig. 8.—The same × 600, about. Sclerites forming labial trough omitted.

Fig. 9.—Ventral external fringe or lip of the labella (left half) × 570.

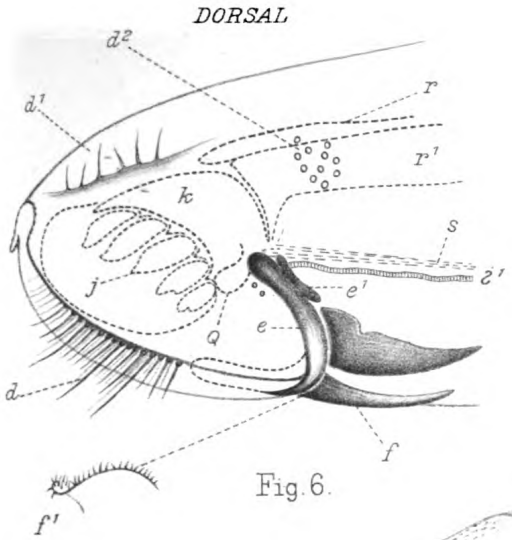


Fig.6.

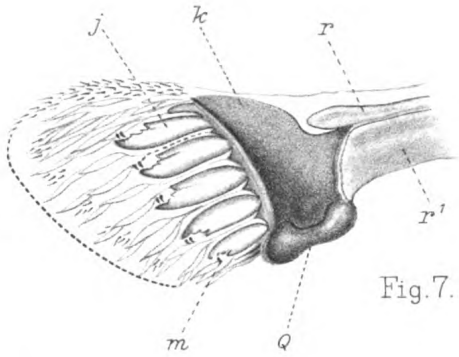


Fig.7.

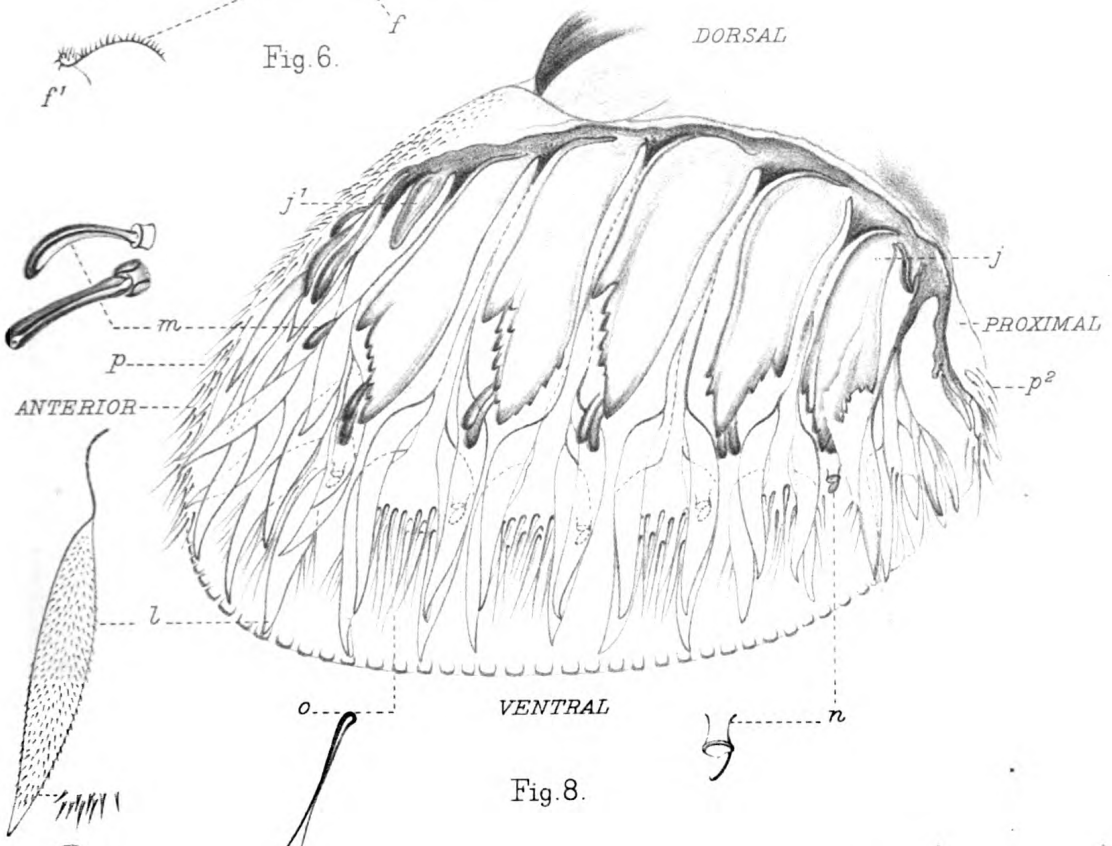


Fig.8.

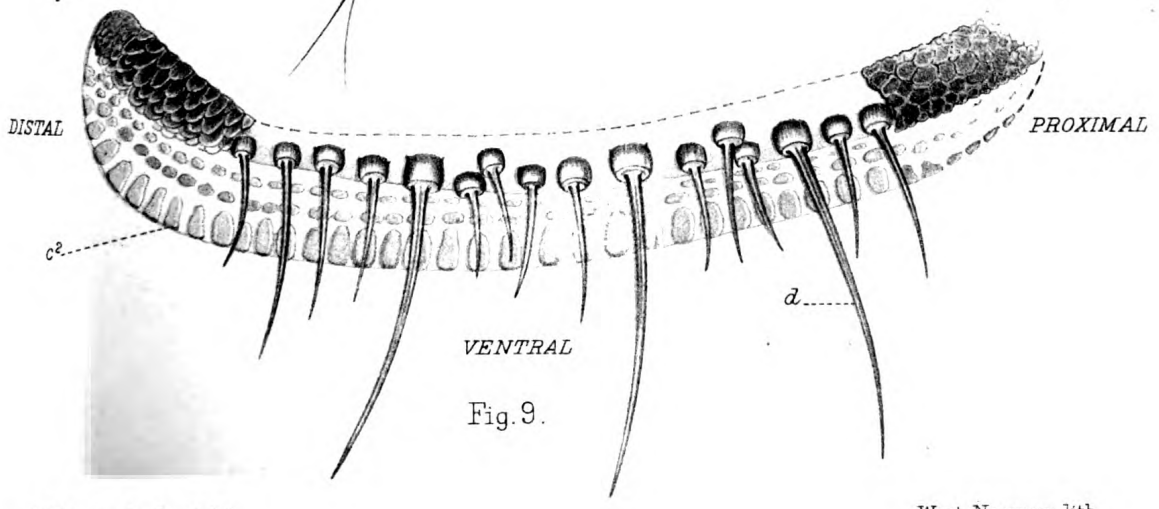


Fig.9.

EXPLANATION OF PLATE XIV

Fig. 10.—Section through everted labella in the plane where the tip of the axial apophysis (q) first appears. The approximate extent of the petiolated blades is shown in dotted lines. × 570.

Fig. 11.—Section through everted labella in the plane where the base of the axial apophysis (q) becomes fused to the tooth-plate (k) and where the labial gutter (r^1) and the dorsal hooked sclerites (r) first appear. × 570.

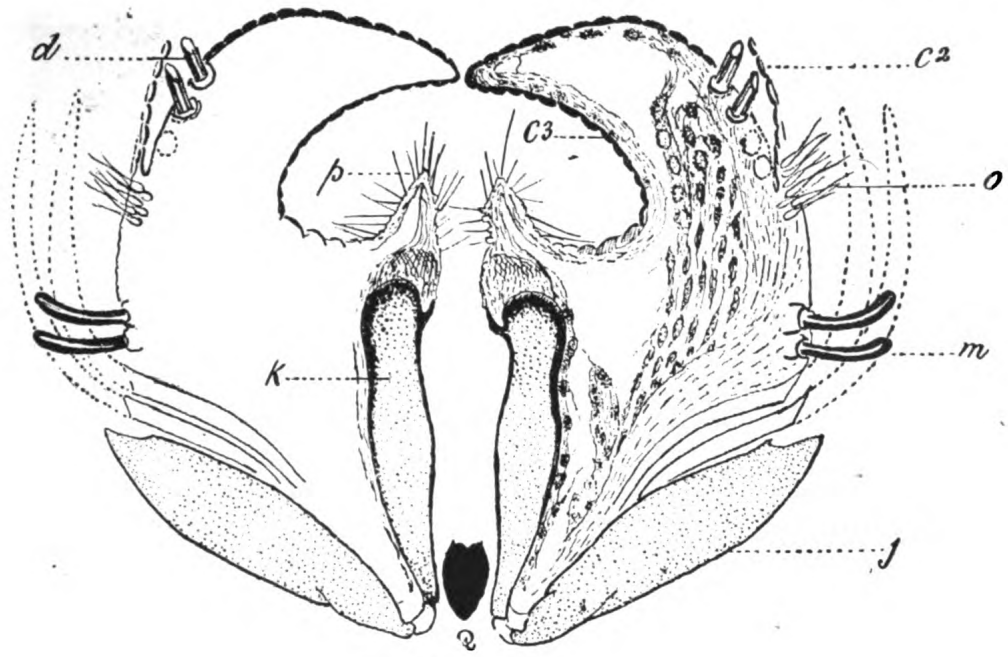


Fig. 10

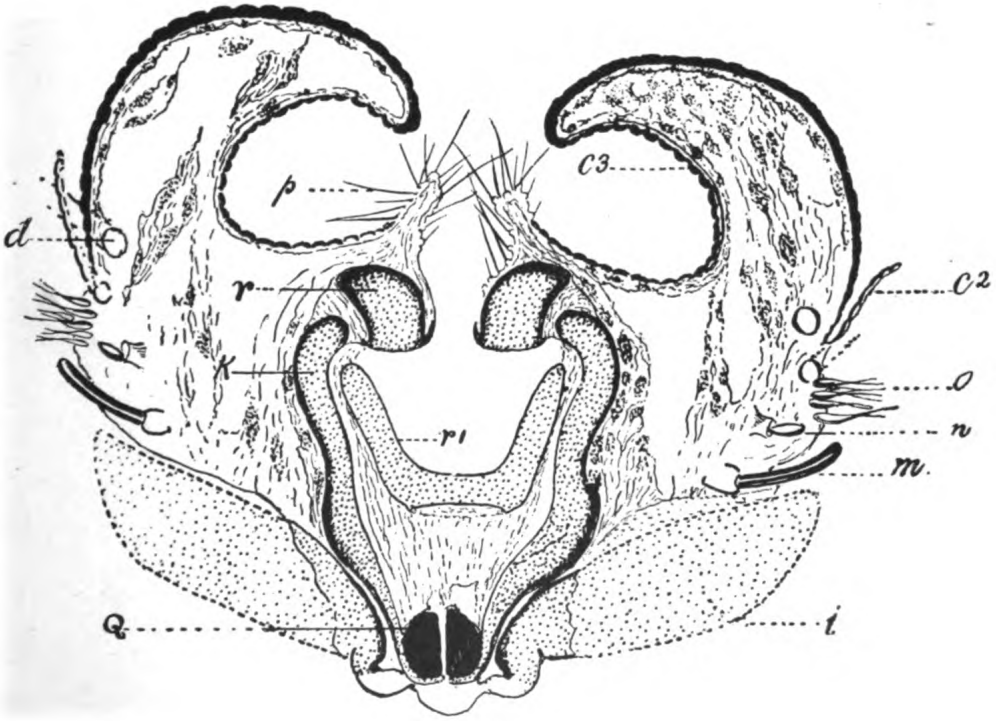


Fig. 11

R. Newstead, ad. nat. del.

EXPLANATION OF PLATE XV

Fig. 12. Section through everted labella in the plane of the fork (e) where the dorsal and ventral folds of the epidermis (c^4-c^7) are well marked. In the next proximal section the tips of the fork are seen to extend into the angle of the third fold (c^6), here indicated by dotted lines. Note that the tooth plate has entirely disappeared and the labial gutter (r, r^1) is now well formed, though the upper sclerites are still free. The approximate plane of this section is between c^6 and e in fig. 14. × 570.

Fig. 13. Pocket formed by the infolding of the epidermis in the plane near d^2 in fig. 14. Here the upper sclerites (r) of the labial gutter have become fused with the lower (r^1). × 570.

Fig. 14. Dorsal aspect of everted labella and distal portion of labium, from a well cleared preparation. Compare this with fig. 2, which was drawn from a preparation which had not been sufficiently cleared.

Fig. 15. Distal portion of labrum. × 200.

Fig. 16. Extreme tip of same showing the scaly dorsal structure. × 570.

Fig. 17. Section through tip of labrum. × 570.

Fig. 18. Section of labrum through the plane just in front of the ventral thickening (fig. 15, a^1) carrying minute hairs. × 750.

Fig. 19. Section of labrum through ventral thickening (fig. 15, a^1) showing minute hairs. × 750.

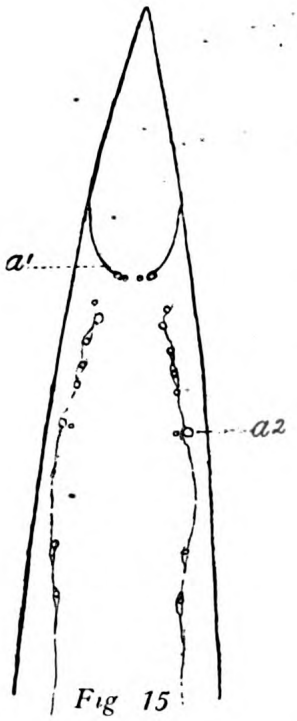
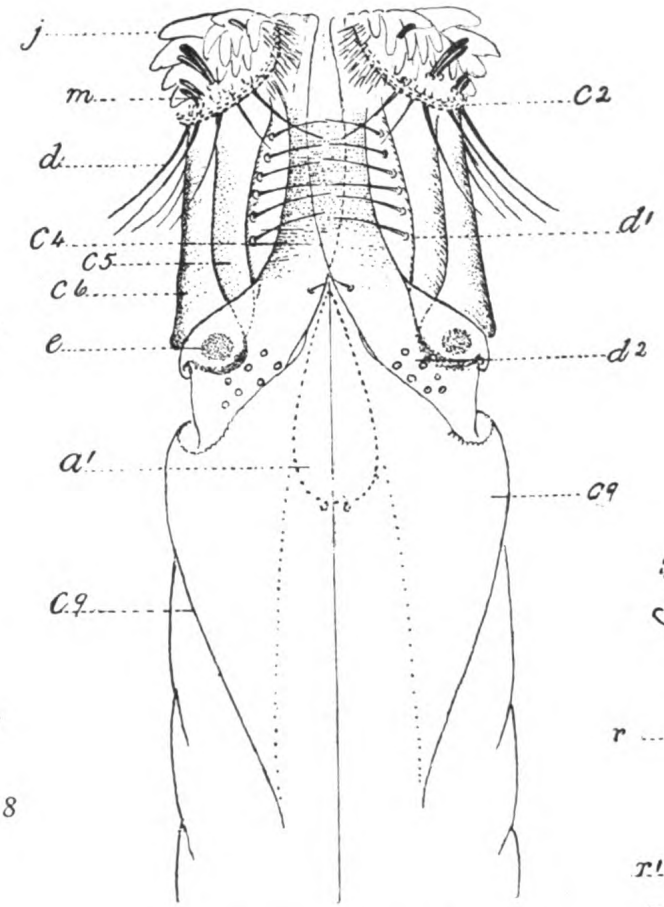


Fig 15

Fig. 14



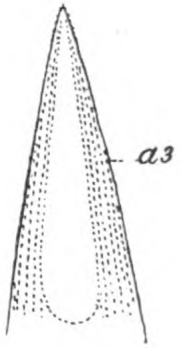


Fig. 16



Fig. 17



Fig 18

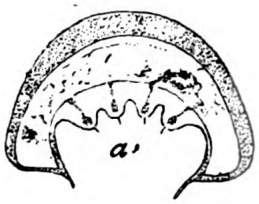


Fig 19

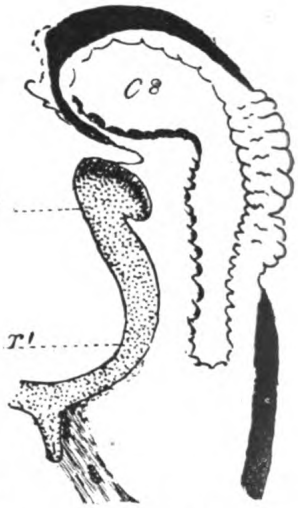


Fig. 13

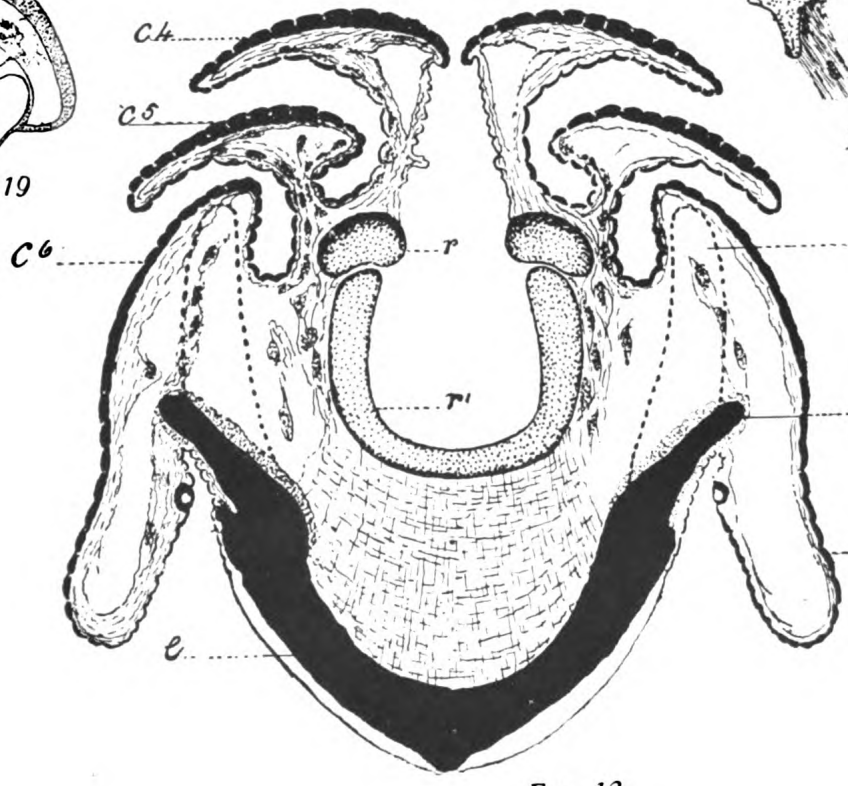


Fig. 12

R. Newstead, ad. nat. del.

EXPLANATION OF PLATE XVI

Fig. 20. Transverse section through the tip of the closed labella. × 600.

Fig. 21.- Section through the closed labella at the plane where the teeth (*j*) have almost disappeared, and where the sclerite forming the labial gutter (r^1) and the tips of the fork (*e*) first appear. × 750.

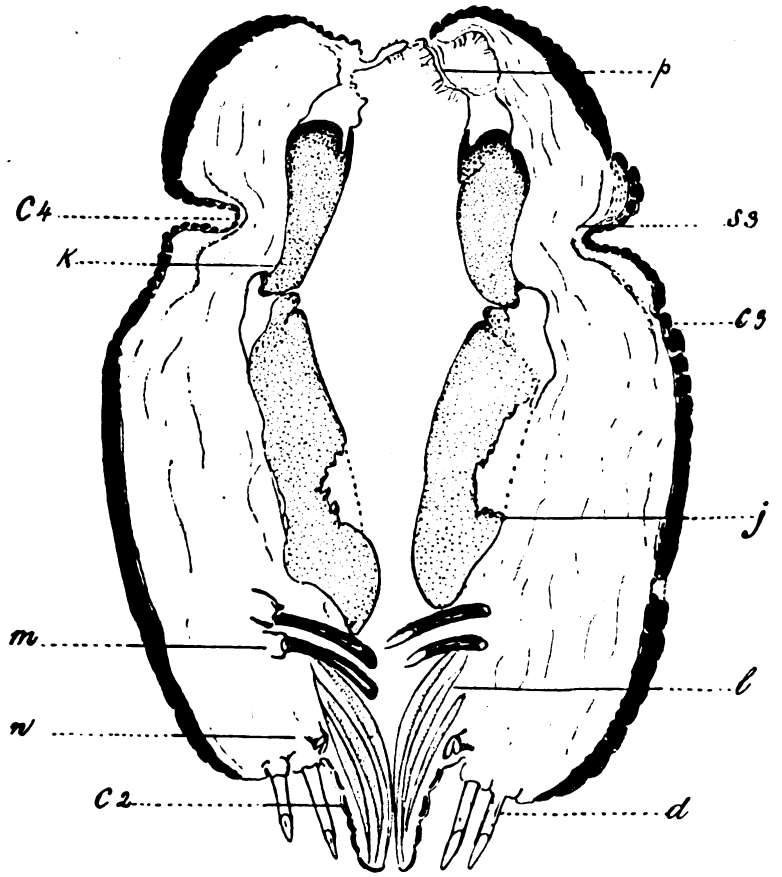


Fig 20

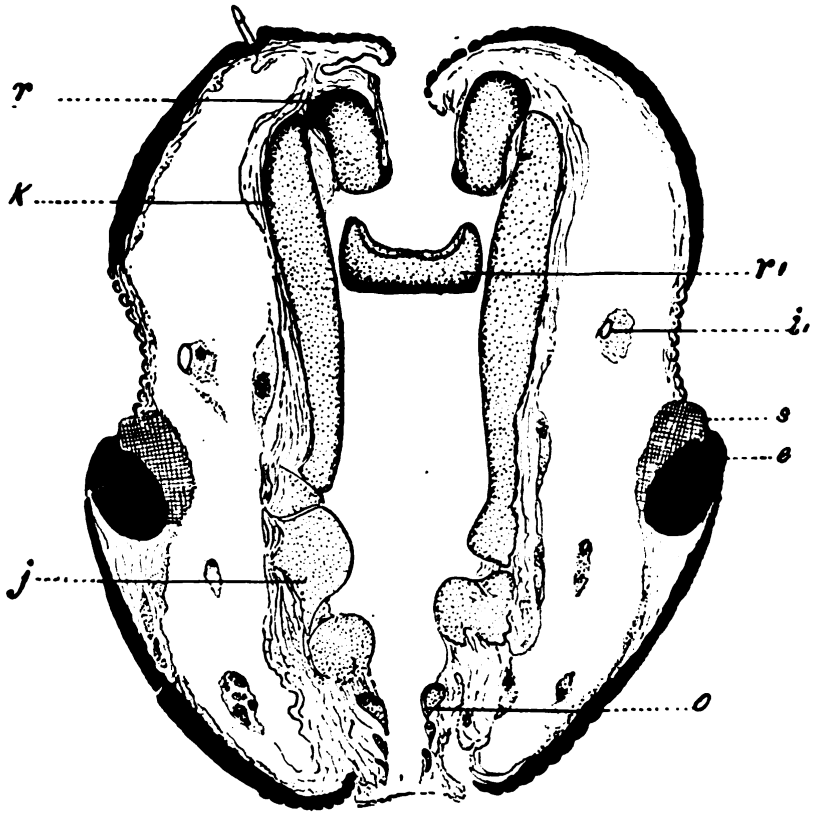


Fig 21

EXPLANATION OF PLATE XVII

Fig. 22. Section through the closed labella in the plane where the free end of the axial apophysis (*q*) first appears. In this section the teeth have disappeared and the labella is closed ventrally. × 750.

Fig. 23. Section showing the fusion of the axial apophysis (*q*) with the tooth plate (*k*). × 750.

Fig. 24. Another phase of the same. × 750.

Fig. 25. Section through the closed labella in the plane at the base of the axial apophysis (*q*). Here the tooth plate has entirely disappeared; the arms of the fork are seen converging, and stretching from tip to tip is a broad membrane; here also are seen the lateral sclerities of the fork (e^1) articulating with that organ. × 750.

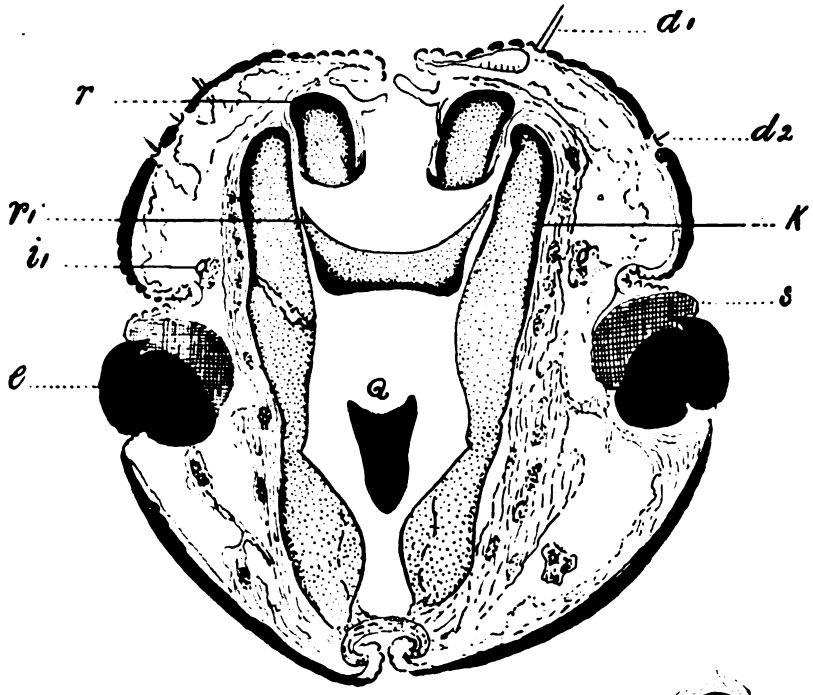


Fig. 22

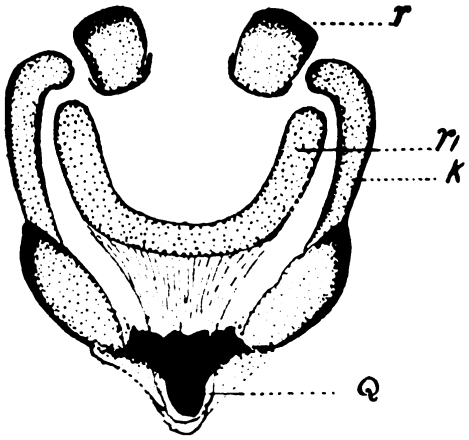


Fig. 23

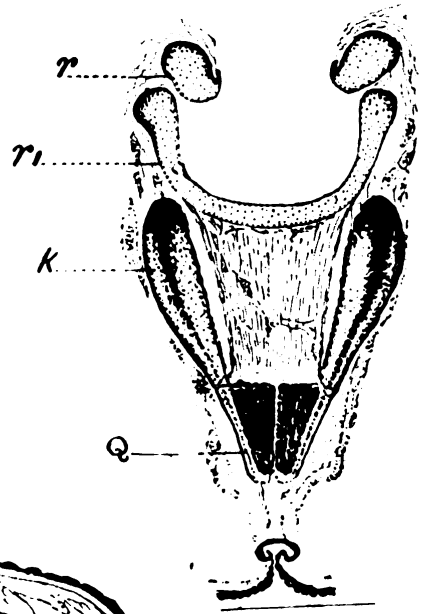


Fig. 24

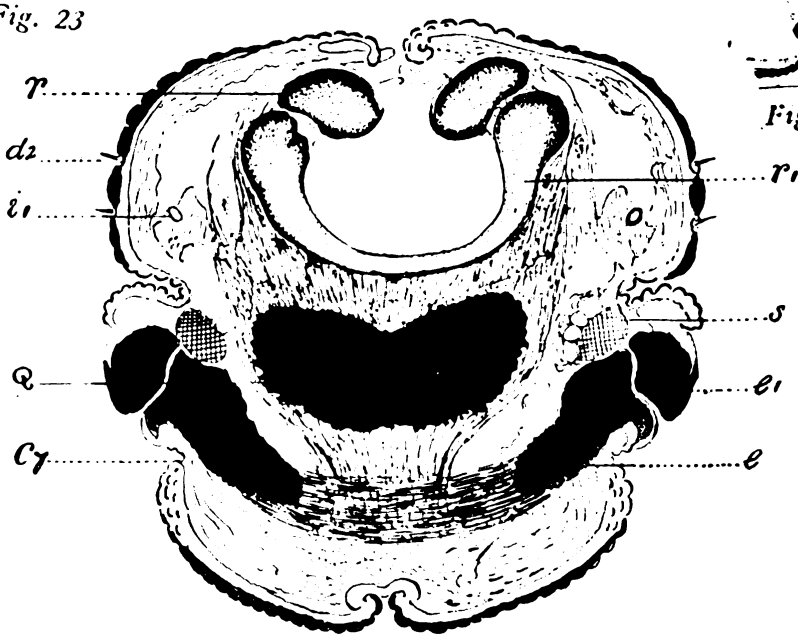


Fig. 25

M

EXPLANATION OF PLATE XVIII

Fig. 26.— Section through closed labella at the junction of the fork (*e*) with the ventral sclerities (*f*). × 750.

Fig. 27.— Section through the labium in the plane where the ventral sclerites (*f*) first appear. The fine hairs at the apex of these organs are here shown. × 750.

Fig. 28.—Section through the labium at the plane where the labrum (*a*) first appears. Here muscular bands stretch across from the ventral sclerites to the walls of the labial gutter. × 150.

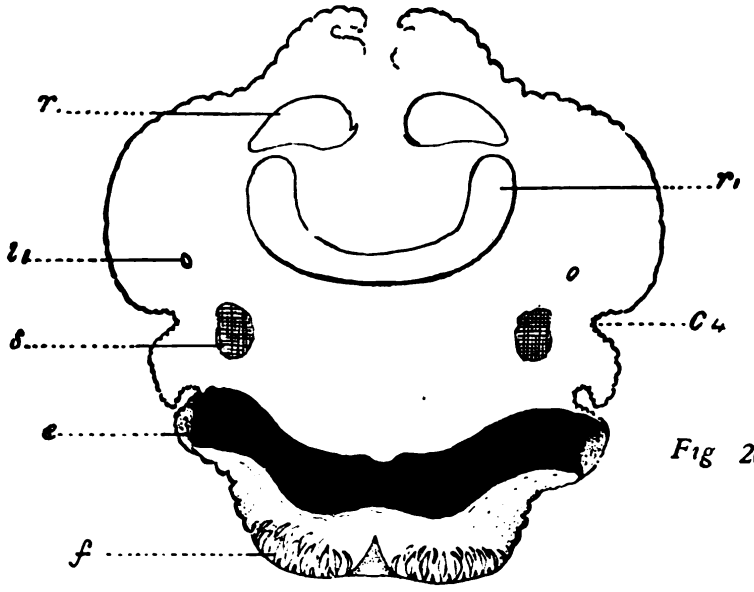


Fig 26

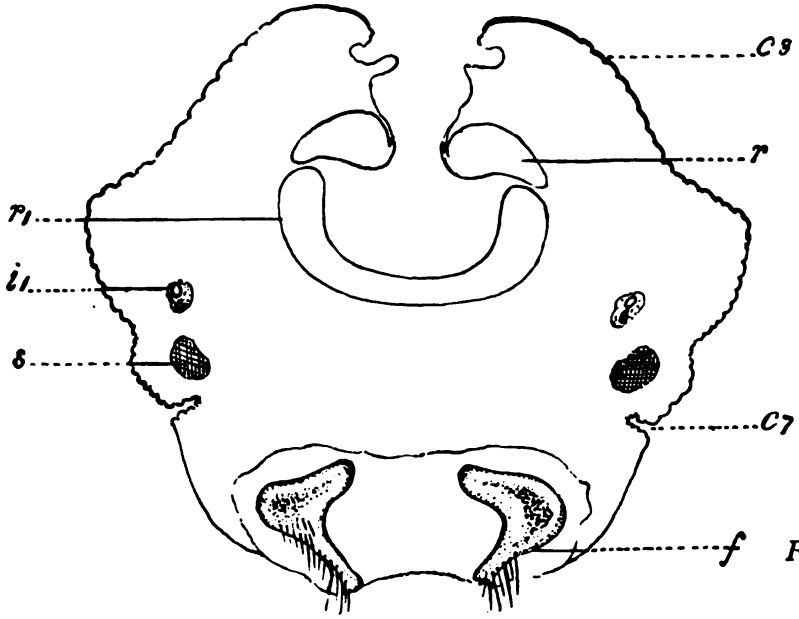


Fig. 27

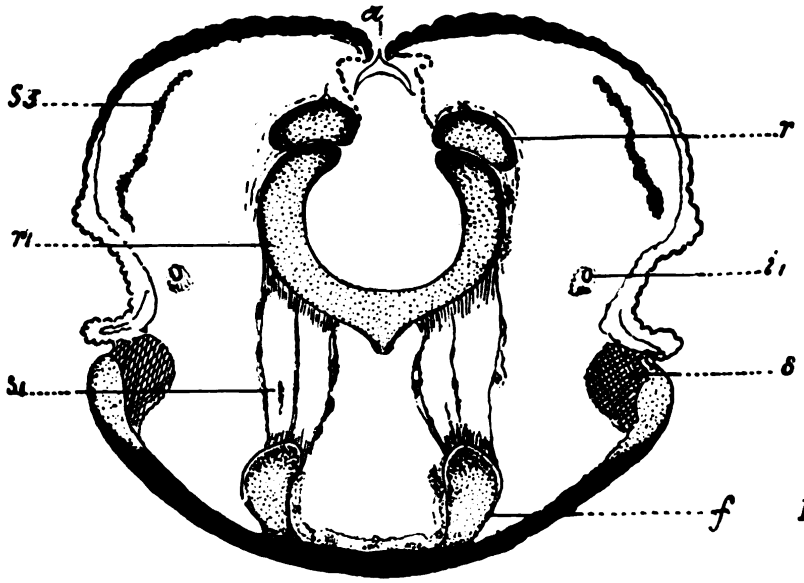


Fig 28

R. Neuwirth ad nat. del.

EXPLANATION OF PLATE XIX

Fig. 29.—Section approximately through the middle plane of the proboscis, in which are seen the labrum (*a*), the hypopharynx (*b*) completely closed and interlocked with the labium. Here also the row-lock like appearance of the labial gutter is well shown and the rod-like sclerites have completely disappeared. × 750.

Fig. 30.—Section through the bulb of the proboscis. Here the hypopharynx is seen fusing with the walls of the labial gutter; and the large air sacs (*i*) have taken the place of the tracheal tubes (fig. 29, i^1, etc.). × 750.

Fig. 31.—Section through the base of the bulbous portion of the proboscis, including the apodemes (*h*) of the labrum. Here the dorsal wall of the hypopharynx has become fused with the labrum, forming the pharyngeal tube. × 200.

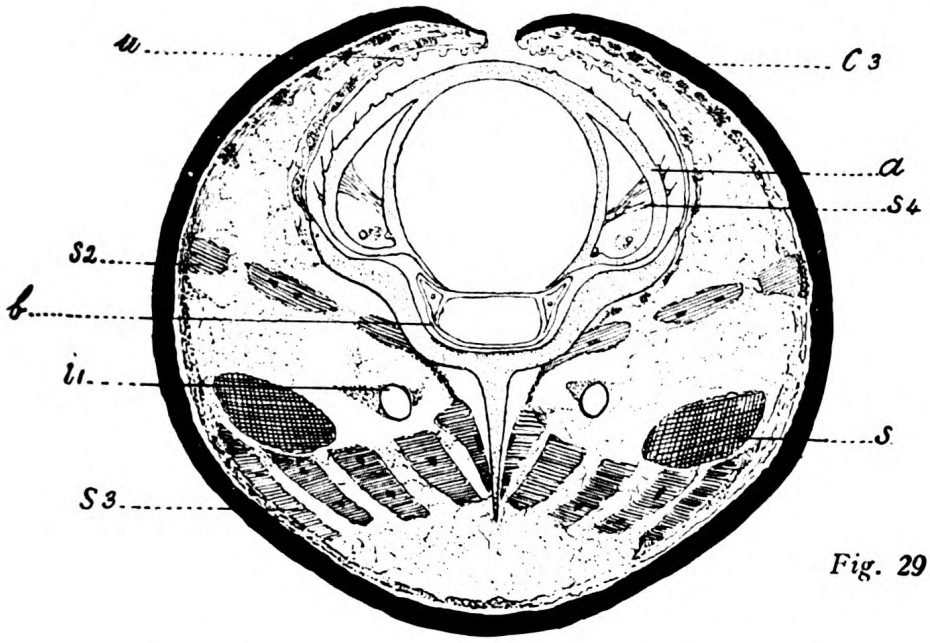


Fig. 29

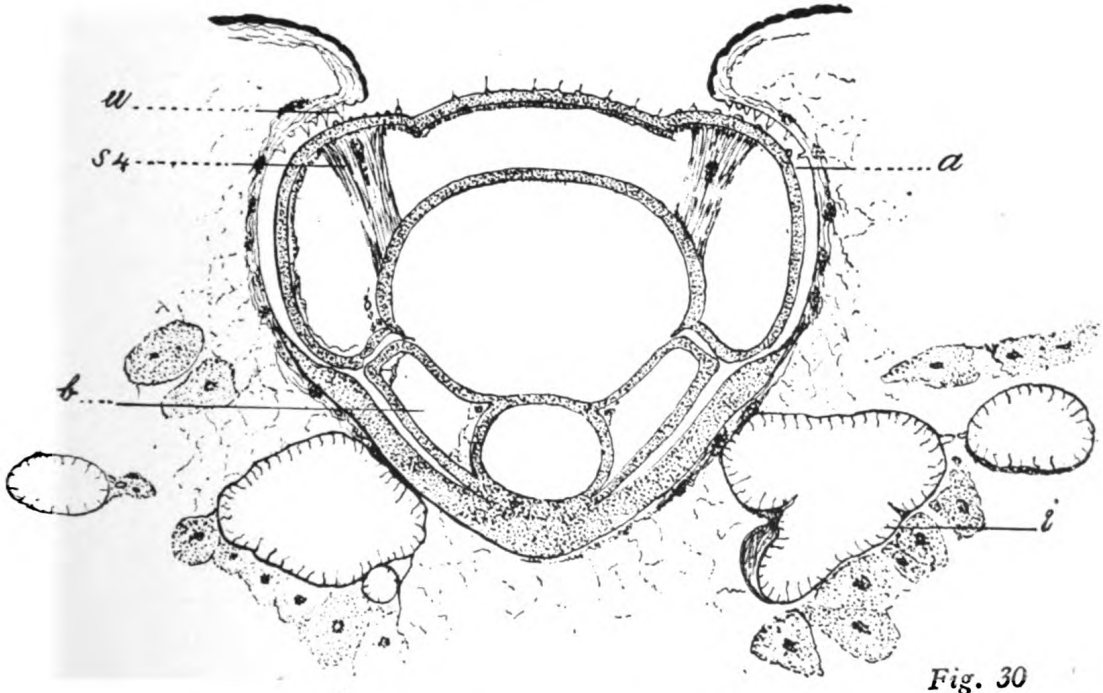


Fig. 30

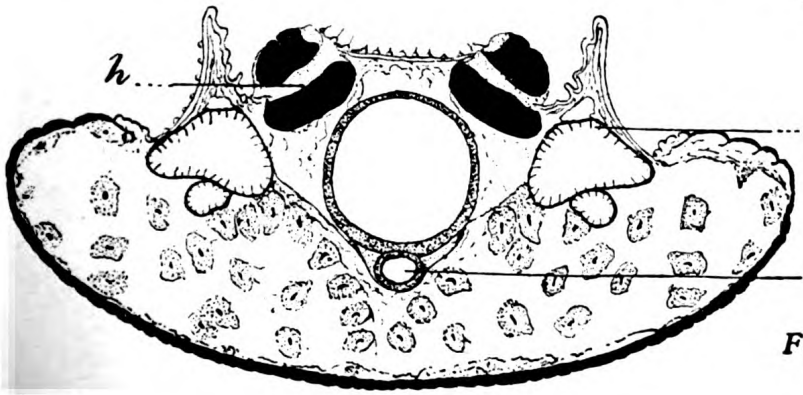


Fig. 31

R. Newstead, ad. nat. del.

TRYPANOSOME TRANSMISSION EXPERIMENTS

TRYPANOSOME TRANSMISSION EXPERIMENTS

BY

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Being the Seventh Progress Report of the Expedition of the Liverpool School of Tropical Medicine to the Congo, 1903-1905

(*Received April 20th, 1907*)

I. ATTEMPTS TO TRANSMIT TRYPANOSOMES BY TSETSE FLIES

In a short series of experiments done in the Gambia in 1902-3,[1] we failed to infect animals with *Trypanosoma gambiense* or *dimorphon* by the bites of captive flies (*Glossina palpalis* and *Stomoxys* (see [1], page 40), which were either freshly caught or had been fed previously on an infected animal. The members of the Royal Society's Commission on Sleeping Sickness in Uganda have since succeeded, however, in infecting healthy animals with *Trypanosoma gambiense* by the bites of flies (*Glossina palpalis*) fed, at the most, 48 hours previously on an infected animal ([2], page 61). As a result of this work it has again been maintained ([13, 17] and others) that the tsetse fly is merely a mechanical transmitter of trypanosomes, and is not an alternative host in which the parasite undergoes definite developmental processes.*

* This suggestion first emanated from Bruce, but he also considered the possibility of there being a developmental process of the trypanosome (*Trypanosoma brucei*) in the tsetse fly (*Glossina morsitans*) (14, page 4).

When our expedition reached the Congo, sleeping sickness was endemic, and the existing conditions were much the same as in Uganda. We therefore determined to repeat the work done there, with the primary object of ascertaining the longest period after feeding on an infected animal at which a tsetse fly, by its bite, is capable of transmitting trypanosomes to a healthy animal. It must be remembered that, like Bruce, we worked with adult flies caught in the bush and that our experimental animals were only partially protected from the chance bites of insects.

The conditions of our experiments were therefore imperfect. The flies used should have been raised in the laboratory from the larvae of known parents. Wild flies, caught when adult in an area where trypanosomiasis is endemic, may have fed when free on infected animals; it is only in flies bred in the laboratory that the dates of all infecting feeds can be accurately known. It is possible that the progeny of infected tsetse flies are capable, or are alone capable,[3] of transmitting the trypanosomes; these points can only be decided by experiments made with laboratory-bred flies.*

We do not think that the danger of chance infection of laboratory animals, only partially protected from the bites of insects, is so great as might be imagined. None of our large stock of rabbits, guinea-pigs, rats and monkeys were ever found to have been so infected during 33 months spent on the Gambia and on the Congo.

In the Congo two main series of experiments were made with tsetse flies, one at Leopoldville and the other at Kasongo. A small "River series" of experiments (Experiments 128, 128a, 129 and part of 211) was done with flies caught along the Congo between Leopoldville and Coquilhatville.†

At Leopoldville it was very difficult to obtain monkeys and we were forced to use guinea-pigs and rats in some of the experiments, although they are, as a rule, rather less susceptible to *Trypanosma gambiense*.[4]

* As is suggested by our notes on the bionomics of captive tsetse-flies (5), it is difficult to breed them. Less than half a dozen of the pupae obtained from the flies used in our experiments in the Congo developed in the laboratory into adult flies.

† For maps illustrating the area traversed by the Expedition and showing the distribution of Sleeping Sickness in the Congo Free State, see Memoir XVIII of the Liverpool School of Tropical Medicine, and (5).

In the Gambia we used small gauze cages and found it difficult to keep tsetse flies (*Glossina palpalis*) alive for more than a few days in captivity. At Leopoldville we attempted to keep them longer by more nearly reproducing their natural habitat in their cages. Cubical gauze cages, measuring 18 inches along each side and containing water and growing grass, were placed in a sheltered position out of doors. At first the experimental animals were simply turned loose in these cages, but the flies were found to feed much better on them if they were immobilized. Under these circumstances flies often lived in captivity for a couple of months. The method, however, had distinct disadvantages: it was difficult to feed the flies on large animals and, because of the size and contents of the cages, it was almost impossible to count the number of living flies they contained.[5] For this reason, in the experiments done at Kasongo the smaller cages were again employed.

Flies were fed on the experimental animals daily.

As a rule, hosts, animals or men, whose blood contained many trypanosomes were chosen for the infecting feeds. In the latter experiments done at Kasongo, when it was found that so few of the previous experiments had been successful, the flies were fed on advanced cases of sleeping sickness, in whose blood there were very few or frequently no parasites, in order that flies should be experimented with which had fed on persons at each stage of the disease. The infecting hosts have been further described in the reports of the successful experiments.

No conclusions concerning the infectivity of the parasites at various stages of the disease can be drawn from an examination of the infecting feeds given to flies used in successful experiments, because other animals remained uninfected after having been fed upon by the same flies under identical conditions.

So far as the conditions permitted, the flies were used for several of the experiments. For example, many of the flies used in Experiment 48 (*To infect a healthy animal by the bites of tsetse flies fed twelve, or more, hours previously on an animal infected with* **Trypanosoma gambiense**) had been used before in other experiments, and many of them had therefore fed several times, but more than 12 hours previously, on infected animals. As far as we know, only *Glossina palpalis* was used in the experiments made at Leopoldville.

At Kasongo about a score of *Glossina fusca* in all were caught. Most of them were allowed to remain in the cages. Where it is necessary experiments on which they fed are specified. Rare examples of *Glossina pallidipes* or *Glossina maculata* may have been present, undetected in the cages, at both localities.*

At Leopoldville *Trypanosoma gambiense* was alone used; at Kasongo *Trypanosoma dimorphon* was also experimented with.

I. FRESHLY-CAUGHT FLIES

To infect susceptible animals by the bites of **Glossina** *caught in localities where trypanosomiasis is endemic.*

There was a great deal of sleeping sickness in the areas where the flies were caught at both Leopoldville and Kasongo. Because of the larger local population, the chances that a newly-caught fly had previously fed on a person infected with trypanosomes were rather greater at Leopoldville. The flies fed at Mswata and Coquilhatville on Experiments 128, 128a, 129, 141 and 211 were caught in very restricted areas where there were many people, the great majority of whom were infected with *Trypanosoma gambiense.* At both Leopoldville and Kasongo trypanosomiasis of animals was far from rare. Because of the presence of a herd of 280 cattle (about 5 per cent. infected) and of considerable game at Kasongo, the chances that a fly caught there had previously fed on an infected animal were much greater than at Leopoldville.

LEOPOLDVILLE.

EXPERIMENT 45.—Guinea-pig. From December 8, 1903, to April 13, 1904, 457 flies were fed. The animal's blood was examined regularly until October, 1904;† trypanosomes were never seen. It died on October 28, 1905, from other causes. No trypanosomes or signs of trypanosomiasis were seen at the autopsy.‡

* See (5) page 63.

† As a routine, the blood in all these experiments was examined, as a rule, two or three times a week, fresh in ¼-inch square coverslip preparations. In addition, the blood of each animal was occasionally centrifugalised (6).

‡ Although it is not always mentioned, in the following experiments an autopsy was done in every case seen within a few hours or minutes of the animal's death, and fresh preparations of body fluids and of organ juices were always examined. By "signs of trypanosomiasis" are meant in particular, enlarged spleen and enlarged, often much congested or haemorrhagic, lymphatic glands.

EXPERIMENT 46.—Guinea-pig. From December 8, 1903, to June 9, 1904, 552 flies were fed. The animal's blood was examined regularly until October, 1904; it died on June 3, 1905, of pneumonia. No trypanosomes or signs of trypanosomiasis were seen at the autopsy.

RIVER SERIES.

EXPERIMENT 128a.—Monkey (*Cercopithecus schmidti*).* On June 24, 25, 26, 1904, 25 flies caught at Mswata were fed on this animal. Its blood was examined regularly until September, 1904, when it died of dysentery; trypanosomes were not seen (malaria present).

EXPERIMENT 129.—Monkey (*Cercopithecus schmidti*). From June 27 to July 7, 1904, 262 flies caught at Mswata and at the sleeping sickness hospital at Coquilhatville were fed on this animal. Its blood was regularly examined, but trypanosomes were never seen; its temperature was always normal. It died (cause ?) on July 9, 1904; no trypanosomes or signs of trypanosomiasis were seen at an autopsy done four hours after death, and a rat sub-inoculated with its blood never became infected.

EXPERIMENT 141.—Monkey (*Cercopithecus schmidti*). From July 15 to July 17, 1905, 52 flies collected at the Coquilhatville sleeping sickness hospital fed on this animal. Its blood was regularly examined until October 14, 1905, when it was stolen. Trypanosomes were never seen in its blood, and its temperature was never abnormal.

KASONGO.

EXPERIMENT 182.—Monkey (*Cercopithecus schmidti*). From January 19 to February 7, 1905, 211 flies fed on this animal. Its blood was frequently examined until July 24, 1905, when it was accidentally killed. No trypanosomes were ever seen (malaria present) during life, and no trypanosomes or signs of trypanosomiasis were present at the autopsy done three hours after death.

Successful Experiment

EXPERIMENT 198.—Monkey (*Cercopithecus schmidti*). From February 8 to April 18, 1905, 2,659 flies fed on this animal. Trypanosomes, probably *Trypanosoma gambiense*, were first seen in its blood on May 13, 1905. It gradually lost weight and died of the disease in November, 1905. In March, two *Glossinae fuscae* fed on this animal.

EXPERIMENT 203.—Monkey (*Cercopithecus schmidti*). From February 22 to March 15, 1905, 1,789 flies fed on this animal. It was accidentally killed on March 16. Its blood was regularly examined; no trypanosomes were seen during life or at the autopsy.

EXPERIMENT 213.—Monkey (*Cercopithecus schmidti*). From March 20 to April 20, 1905, 717 flies were fed. The blood was carefully examined up to July 5, when the animal was killed by its cage companions. No trypanosomes were seen during life or at the autopsy; nor were there any signs of trypanosomiasis. Two puppies sub-inoculated at the autopsy with respectively 3 and 5 c.cm. of blood never became infected.

* Each species of monkey used in these experiments has been shown to be easily infected by *Trypanosoma gambiense* by the inoculation of blood or cerebro-spinal fluid from sleeping sickness patients.

II. INTERRUPTED FEEDING

To infect susceptible animals with **Trypanosoma gambiense** *by the bites of tsetse flies fed immediately previously on an infected animal or man.*

LEOPOLDVILLE.

Successful Experiment

EXPERIMENT 63.—Guinea-pig. This animal was immobilized and placed together with an infected guinea-pig in a cage containing tsetse-flies. From December 24, 1903, to January 15, 1904, 219 flies fed on one or both animals. Trypanosomes appeared in the blood of this animal on January 15, 1904, and it died of the disease (and other complications) on March 16, 1904. The morphology and animal reactions of the parasites agreed with *Trypanosoma gambiense*.

The flies used in this experiment were infected by feeding on guinea-pigs inoculated directly from an early case of trypanosomiasis. Trypanosomes were very numerous in their peripheral blood, and they had been infected for about two months.

EXPERIMENT 76.—Monkey (*Colobus satanas*). Flies in a small cage were allowed to partially fill themselves with blood from a "sleeping sickness" patient, and were then permitted to finish their feed on this monkey. From January 2 to April 12, 1904, when the monkey died of sunstroke, 147 flies were fed in this manner. Trypanosomes were never seen in the monkey's blood and its temperature remained normal.

EXPERIMENT 87.—Guinea-pig. As in Experiment 63, this animal was placed with an infected one in a cage containing tsetse-flies. From January 23 to March 8, 1904, 372 flies were fed. Its blood was examined regularly until June 7, and from then at intervals until it died of exposure in October, 1905. Trypanosomes were never seen; there were no signs of trypanosomiasis at the autopsy.

KASONGO.

EXPERIMENT 209.—Monkey (*Cercopithecus schmidti*). From March 3 to April 18, 1905, 183 flies completely filled themselves with blood from this animal after having commenced immediately previously to feed on an infected animal. Its blood was examined daily, but trypanosomes were never seen. The animal died on April 24 (cause, ? starvation). No trypanosomes or signs of trypanosomiasis were seen at the autopsy done immediately after death.

III. THREE TO FIVE HOURS INTERVAL

To infect a susceptible animal with **Trypanosoma gambiense** *by the bites of tsetse flies fed from 3 to 5 hours previously on an infected animal.*

Very few flies were used in these experiments, since it was exceedingly difficult to get them to feed at such a short interval.

EXPERIMENT 205.—Monkey (*Cercopithecus schmidti*). From February 24 to March 10, 1905, 44 flies were fed. Blood was examined for trypanosomes without result (malaria present) until the animal died of dysentery on April 17. No trypanosomes or signs of trypanosomiasis were seen at the autopsy.

EXPERIMENT 231.—Monkey (*Cercopithecus schmidti*). On April 24, 25, 26, 1905, 22 flies were fed. The blood was regularly examined up to November, 1905. Trypanosomes were never seen. The animal died of dysentery on February 21, 1906; no trypanosomes or signs of trypanosomiasis were seen at the autopsy.

IV. EIGHT TO TWELVE HOURS INTERVAL

To infect a susceptible animal with trypanosomes by the bites of tsetse flies fed at least 8 to 12 hours previously on an infected man or animal.

LEOPOLDVILLE.

Trypanosoma gambiense

EXPERIMENT 88.—Monkey (*Cercopithecus schmidti*). From January 27 to May 28, 1904, 634 flies, previously fed on a sleeping sickness patient, fed on this animal. Its blood was examined frequently until March 15, 1904, and then at intervals until it was killed by rats on March 30, 1905. Its temperature was always normal and trypanosomes were never seen in its blood.

RIVER SERIES.

EXPERIMENT 128. Monkey (*Cercopithecus schmidti*). From June 24 to July 22, 1904, 87 flies, previously fed on a monkey infected with *Trypanosoma gambiense*, were allowed to feed on this animal. Its blood was examined at intervals until September 28, 1904; trypanosomes were never seen. The temperature-chart was not characteristic of trypanosomiasis (malaria present).

KASONGO.

(*a*) *Trypanosoma gambiense*

EXPERIMENT 187.—Monkey (*Cercopithecus* sp. ?). From January 28 to April 10, 1905, 939 flies, previously fed on a sleeping sickness patient, fed on this animal. Its blood was examined almost daily until May 5, 1905, when it was stolen; trypanosomes were never seen.

EXPERIMENT 197. Monkey (*Cercopithecus mona*). From February 9 to April 20, 1905, 943 flies, previously fed on infected guinea-pigs, were fed on this animal. Its blood was examined regularly until it died on July 6 (on river steamer) from exposure. Trypanosomes were never seen (malaria present) during life, or at the autopsy done immediately after death.

(*b*) *Trypanosoma dimorphon*

EXPERIMENT 216.—Monkey (*Cercopithecus mona*). From March 25 to April 25, 1905, 388 flies were fed. Blood was regularly examined until June 13, when the animal was moribund. It was therefore killed. Trypanosomes were never seen during life, and no signs of trypanosomiasis were seen at the autopsy. (Cause of death ? exposure).*

* During the journey of the expedition, on foot, from Kasongo to Lusambo (5), the monkeys unavoidably received considerable hard usage.

V. TWELVE TO FIFTEEN HOURS INTERVAL

To infect a susceptible animal with **Trypanosoma gambiense** *by the bites of tsetse flies fed at least 12 to 15 hours previously on an infected animal or man.*

LEOPOLDVILLE.

EXPERIMENT 48.—Guinea-pig. From December 12, 1903, to April 6, 1904, 1,706 flies were fed. The blood was regularly examined until April 12, 1904, when the animal died. Trypanosomes were never seen in its blood nor in any of the body-fluids or organ-juices examined at the autopsy, done immediately after death.

EXPERIMENT 61.—Rat. From December 23, 1903, to May 20, 1904, 484 flies were fed on this animal. Its blood was regularly examined until July 3, 1904, when it was accidentally killed. Trypanosomes were not seen.

EXPERIMENT 62.—Rat. From December 23, 1903, to February 25, 1904, 262 flies fed on this animal. No trypanosomes were ever seen in its blood. It died on March 1, 1904, of a lung disease.

EXPERIMENT 71.—Rat. From December 18, 1903, until May 15, 1904, 417 flies were fed on this animal. Its blood was regularly examined for trypanosomes without result until July 7, 1904, when it was accidentally killed.

In the four preceding experiments it was attempted to transmit the trypanosomes from animal to animal; in the three following it was attempted to transmit them from man to animal.

EXPERIMENT 83.—Rat. From January 16 to April 9, 1904, 999 flies were fed on this animal. Its blood was regularly examined for trypanosomes with negative result until July 1, 1904, when it was accidentally killed.

EXPERIMENT 107.—Guinea-pig. From April 13 to May 30, 1904, 390 flies were fed on this animal. Its blood was examined regularly until June 1, and afterwards at intervals until October 8, when, since trypanosomes were still absent from its blood, observations were discontinued. This animal was afterwards successfully inoculated with *Trypanosoma dimorphon* in experiments made to ascertain the animal reaction of a Congo cattle trypanosome.

KASONGO.

EXPERIMENT 186.—Monkey (*Cercopithecus* sp. ?). From January 24 to April 5, 1905, 542 flies, previously fed on a sleeping sickness patient, fed upon this animal. Its blood was examined frequently until it was stolen on May 4; trypanosomes were never seen.

EXPERIMENT 206.—Monkey (*Cercopithecus schmidti*). From February 25 to April 20, 1905, 468 flies, previously fed on an infected animal, were fed on this monkey. On April 25 it was badly injured by its cage companions, and was therefore killed. No trypanosomes were seen during life (malaria present), or at the autopsy, nor were there any signs of trypanosomiasis.

VI. FIFTEEN TO TWENTY HOURS INTERVAL

To infect a susceptible animal with **Trypanosoma gambiense** *by the bites of tsetse flies fed from 15 to 20 hours previously on an infected animal.*

LEOPOLDVILLE.

EXPERIMENT 97.—Guinea-pig. From February 20 to May 16, 1904, 395 flies were fed. Its blood was regularly examined for trypanosomes without success until May 16, when it died from a ruptured stomach. No trypanosomes or signs of trypanosomiasis were seen at the autopsy.

VII. EIGHTEEN TO TWENTY-FOUR HOURS INTERVAL

To infect a susceptible animal with **Trypanosoma dimorphon** *by the bites of tsetse flies fed at least 18 to 24 hours previously on an infected animal.*

KASONGO.

EXPERIMENT 212.—Domestic cat. From March 13 to April 29, 1905, 510 flies fed. Blood was examined regularly until July 26, 1905. Trypanosomes were never seen. When examinations were recommenced in September, 1905, trypanosomes were present. The animal died in November, 1906, of the disease. In May one *Glossina fusca* fed on this animal; a single *Glossina pallidipes* also had an opportunity of feeding. The flies used were infected by feeding on three guinea-pigs inoculated about two months previously with blood from a horse naturally infected with *Trypanosoma dimorphon*; they frequently fed when the parasites were either absent or very scanty in the peripheral blood. **Successful Experiment**

VIII. TWENTY-FOUR HOURS INTERVAL

To infect a susceptible animal with **Trypanosoma gambiense** *by the bites of tsetse flies fed at least 24 hours previously on an infected animal or man.*

KASONGO.

EXPERIMENT 208.—Monkey (*Cercopithecus schmidti*). From March 3 to April 28, 1905, 590 flies, previously fed on an infected animal, were fed on this monkey. On June 16, trypanosomes were first seen in its blood. It died on October 2, 1905, from pneumonia. The spleen was considerably enlarged. No trypanosomes were seen at the autopsy, and none had been present in the blood for the preceding six weeks. In March, 3 *Glossina fusca* had opportunities for feeding on this animal. All the flies used in this experiment were infected by being fed on a guinea-pig, whose blood contained very numerous trypanosomes, inoculated three months previously with blood from a case of sleeping sickness. Many of these flies had fed three or more days previously on sleeping sickness cases whose blood contained very few or no trypanosomes. **Successful Experiment**

EXPERIMENT 218.—Monkey (*Cercopithecus schmidti*). From April 3 to April 14, 1905, 71 flies, previously fed on a sleeping sickness patient, were fed on this monkey. The blood was examined at intervals until June 5; trypanosomes were never seen. The animal was moribund on June 5; it was therefore killed and an autopsy was made immediately. No trypanosomes or signs of trypanosomiasis were observed.

IX. FOUR DAYS INTERVAL

To infect a susceptible animal with **Trypanosoma gambiense** *by the bites of tsetse flies fed at least four days previously on a sleeping sickness patient.*

LEOPOLDVILLE.

EXPERIMENT 74.—Guinea-pig. From December 30, 1903, to May 29, 1904, 425 flies were fed on this guinea-pig. Its blood was examined regularly until October, 1904, when it died of coccidiosis. No trypanosomes were ever seen.

EXPERIMENT 122.—Monkey (*Cercopithecus schmidti*). From April 23 to June 4, 1904, 209 flies were fed on this monkey. Its blood was examined regularly until June 14, when it was accidentally killed. Its temperature had always been normal, and at the autopsy, as during life, trypanosomes were not seen.

It was thought that perhaps the trypanosomes underwent a lengthy developmental process in the tsetse fly, and that therefore the scanty success of the preceeding experiments might have been due to the shortness of the interval between the infecting and transmitting feeds. In the two following sets of experiments this period was consequently greatly increased.

X. TEN DAYS INTERVAL

To infect a susceptible animal with **Trypanosoma dimorphon** *by the bites of tsetse flies fed at least ten days previously on an infected animal.*

KASONGO.

EXPERIMENT 215.—Guinea-pig. From March 23 to April 25, 1905, 210 flies fed; in many instances 35 days intervened between the first infecting feed and the transmitting feed. The blood was examined regularly until August 4; no trypanosomes were ever seen. Examinations were then discontinued until September 18, 1905, when the animal died of tuberculosis. No trypanosomes were seen at the autopsy.

EXPERIMENT 217.—Monkey (*Cercopithecus schmidti*). From April 2 to April 27, 1905, 206 flies were fed; in many instances 25 to 30 days intervened between the first infecting feed and the transmitting feed. The blood was examined regularly up to July 29; no trypanosomes were ever seen (malaria present). The animal died September 29, 1905, of diarrhoea; no trypanosomes or signs of trypanosomiasis were seen at the autopsy done immediately after death.

XI. FOURTEEN DAYS INTERVAL OR MORE

To infect a susceptible animal with Trypanosoma gambiense *by the bites of tsetse flies fed at least fourteen days previously on an infected animal.*

LEOPOLDVILLE.

EXPERIMENT 79.—Guinea-pig. From January 11 to May 24, 1904, 156 flies fed on this animal. Its blood was examined regularly until May 27, and then at intervals until October, 1904. Trypanosomes were never seen. The animal died in November, 1905. No trypanosomes were seen at the autopsy.

XII. MISCELLANEOUS

EXPERIMENT 211.—Guinea-pig. This animal was used for feeding odd batches of tsetse-flies which for various reasons could not be employed in one of the set experiments.

From May 16, 1904, to April 30, 1905, 1,278 flies were fed on it; 200 of these flies were freshly caught, the remaining had been fed at various antecedent periods on animals infected with either *Trypanosoma gambiense* or *Trypanosoma dimorphon*. The blood was examined until August, 1905; trypanosomes were never seen. The animal died in September, 1905 (cause of death ?). No trypanosomes or signs of trypanosomiasis were seen at the autopsy done immediately after death.

In the Gambia it was thought that the excessively dry atmosphere at the season when our experiments were done might have accounted for their failure[1]. This objection cannot be made to the Congo experiments; when they were done, the average morning humidity (6 a.m.) at Leopoldville was about 45 per cent., at Kasongo about 94 per cent.

Experiments 129, 203, 206, in which the animals were not observed for a very long period, should probably be disregarded. The remaining observations should be considered in conjunction with the published reports of similar experiments done in Uganda [8, 10].

Bruce, Nabarro and Greig (8).—These authors publish three experiments in which animals were infected with *Trypanosoma gambiense*, within from two to four and a half weeks from the commencement of the experiment, by the bites of from 200 to 1,000 freshly-caught *Glossina palpalis*. The rapidity with which these experiments succeeded is noteworthy.

In five experiments they succeeded in transmitting *Trypanosoma gambiense* by the bites of from 250 to 570 flies fed from eight to forty-eight hours previously on infected animals. The length of time from the commencement of the experiment to the recognition of the infection varied from seven to ten weeks.

Greig and Gray (10).—In five experiments these authors succeeded in transmitting cattle trypanosomes by the bites of *Glossina palpalis*. The period elapsing between the commencement of the experiment and the detection of the parasites in the blood of the experimental animal varied from four and a half to seventeen and a half weeks; the number of flies fed ranged from 151 to 923.

It is seen that all the results are in conformity with the hypothesis that *Glossina palpalis* transmits *Trypanosoma gambiense* mechanically, and that it is probably not able to do so when the space between transmitting and infecting feeds much exceeds 48 hours. This conclusion is, nevertheless, to our minds a most unsatisfying one if we are to regard this *Glossina* as the chief* or only† carrier of *Trypanosoma gambiense.*

The experiments of all observers show that it is frequently necessary to feed hundreds, almost thousands, of flies on a susceptible animal before it becomes infected.‡

It may be objected that in many of these experiments flies may have been unnecessarily fed on the animals after they were infected but before the infection was recognised. This objection is partially negatived by observations (7, 8, see page 20) showing that the incubation period of a natural infection by *Trypanosoma gambiense* may be so short as from two to four weeks.

If the number of successes obtained in such experiments, where flies are fed on an infected animal whose blood is swarming with parasites, is so small, it scarcely seems possible that mechanical transmission by tsetse flies can alone be responsible for the *rapid* spread of sleeping sickness of recent years. From these experiments it seems that, as a rule, a native must be bitten by a comparatively large number of flies, which have fed not more than 48 hours previously on a case of human trypanosomiasis§ before he will become infected; it must be remembered that trypanosomes are usually rather rare in the blood of cases of sleeping sickness.

* The possibility that varieties of *Glossina* other than *palpalis* may carry *Trypanosoma gambiense* must not be forgotten. It has recently been shown that *Glossina palpalis* may transmit trypanosomes other than *Trypanosoma gambiense*; Greig and Gray have shown that it will convey the trypanosomes found in various domestic animals in Uganda (10). In Experiment 212 of this paper we succeeded in transmitting *Trypanosoma dimorphon* by *Glossina palpalis*. Koch (9) suggests that in German East Africa cattle trypanosomes may be transmitted by *Glossina pallidipes* and *Glossina fusca* as well as by *Glossina morsitans*, which was formerly alone thought to have that power.

† The Sergents (18) transmitted *T. brucei*, previously only known to be carried by *G. morsitans*, with an interval of 48 hours between infecting and transmitting feeds, by the bites of a tabanid (*A. nemoralis*).

‡ Fewer flies are of course needed in an experiment when there is practically no interval between the feeds on the infected and experimental animals.

§ *Trypanosoma gambiense* has as yet been found in nature in no other host than man.—May 7: Since this was written Report VIII of the Royal Society's Commission on Sleeping Sickness has been received. Its authors believe that they have found *T. gambiense* in native dogs.

Although the chronicity of the disease is well known, it seems impossible that large percentages of populations, whose vocation does not keep them constantly on the water, should become infected in such a way in places where tsetse flies are far from plentiful, like Kalombe (17 per cent. infected), Dibwe (18 per cent.), Miambwe (7 per cent.), Lokula (6 per cent.) in the Congo Free State.

It seems certain that such a mechanical transmission cannot be the only way in which *Trypanosoma gambiense* is transmitted from man to man. We believe either that something is wrong in the way in which *Glossina palpalis* has been used in these experiments, or that *Trypanosoma gambiense* can be conveyed by some other means than by it.*

The status of cattle trypanosomiasis (see page 233) in the Congo Free State furnishes an additional objection to explaining the propagation of trypanosomiasis by mechanical transmission alone. A considerable percentage of the cattle are infected. Trypanosomes are very rarely seen in their blood. Enormous quantities (10 to 50 c.cm. or more) of infected blood must be injected into susceptible animals before they become infected, and failures to infect are common. It does not seem possible that the mere mechanical transmission of an infinitesimal quantity of blood from animal to animal by an insect's proboscis can adequately explain the distribution of a strain of trypanosomes possessing so little virulence in direct inoculations.

II. ATTEMPTS TO TRANSMIT TRYPANOSOMES BY VARIOUS BLOOD-SUCKING ARTHROPODS

If the tsetse fly is merely a mechanical transmitter of trypanosomes, there seems no reason why other blood-sucking arthropods should not also transmit them. To test this possibility the experiments described below were made with the "Congo floor maggot" (larva of *Auchmeromyia luteola*), Anophelines (*Pyretophorus costalis*), *Simulium* and *Ornithodorus moubata*. None of them were successful. As before, animals showing large numbers of trypanosomes in their blood were preferred for the infecting feeds.

I. EXPERIMENTS WITH LARVAE OF *AUCHMEROMYIA LUTEOLA*

I. FRESHLY-CAUGHT LARVAE

(*a*) *To ascertain whether larvae caught in native huts in an area where sleeping sickness is endemic contain trypanosomes in a state capable of infecting susceptible animals.*

* *T. equiperdum*, usually only transmitted by coitus, may be spread in a totally different manner by the bites of fleas (25).

EXPERIMENTS 41 and 42.—Rats. On November 20, 1903, these two animals were inoculated subcutaneously with the fluid obtained by rubbing up 16 larvae, freshly-caught at Wathen, in normal saline. Both rats were examined frequently (every three days) until April, 1904; neither ever became infected.

It is noteworthy that no trypanosomes were seen in the blood from the alimentary canals of six freshly-caught larvae, although living trypanosomes have been found in the intestines of larvae up to 10 to 13 hours after they had fed on a known infected animal.

(*b*) *To ascertain whether larvae, caught in native huts at Wathen and Leopoldville, are able to infect a susceptible animal with trypanosomes by their bites.*

The larvae experimented with were always left in tins containing dry sand. Animals, on which it was desired that the larvae should feed, were tied to an arched, wire frame-work and placed in the tins so that their cleanly-shaved abdomens rested on the surface of the sand. Under these conditions, the larvae commenced to feed in about an hour. They apparently fed daily and for many minutes at a time, unless the animals were moved, when they immediately burrowed into the sand.

EXPERIMENT 44.—Guinea-pig. From December 18, 1903, to March 12, 1904, 224 larvae fed. The blood was examined frequently until May, 1904, and then at intervals until November, 1905, when the animal died of pneumonia; trypanosomes were never seen.

II. INTERRUPTED FEEDING

To ascertain whether larvae immediately previously fed on an animal heavily infected with **Trypanosoma gambiense** *were capable of transmitting the parasite by their bites to a susceptible animal.*

In Experiments 107a and 108 an infected guinea-pig was placed for one or two hours in the tin containing the larvae; it was then quickly replaced by a healthy animal which was left in the tin overnight. When the infected animals were removed a dozen maggots were frequently seen to be feeding on them; it therefore seems highly probable that many of these resumed their feed on the second animal. In the records of these experiments the number of larvae which fed during the night on either or both of the animals is noted.

EXPERIMENT 107a.—Guinea-pig. On March 11, 1904, 39 larvae fed on one or both animals. Blood was examined regularly until May, and then at intervals until October, 1904, when this guinea-pig was used for another experiment; trypanosomes were never seen.

EXPERIMENT 108.—Guinea-pig. From March 13 to 27, 1904, 194 larvae were fed. The blood was examined regularly until the end of May, and then at long intervals until September, 1905 (death from pneumonia); trypanosomes were never seen.

III. TWELVE HOURS INTERVAL

To infect a susceptible animal with *Trypanosoma gambiense* by the bites of larvae fed at least 12 hours* previously on an infected animal.

EXPERIMENT 60.—Guinea-pig. From December 30, 1903, to April 27, 1904, 454 larvae were fed. The blood was frequently examined until May, 1904, and then at intervals until October, 1905 (death from pneumonia); trypanosomes were never seen.

II. EXPERIMENT WITH ANOPHELINES

The mosquitoes used in this experiment were bred from larvae caught at Leopoldville; they were practically all *Pyretophorus costalis*.

To infect a susceptible animal with **Trypanosoma gambiense** *by the bites of anopheles fed on an infected animal at least 24 hours previously.*

EXPERIMENT 53.—Guinea-pig. From December 15, 1903, to May 8, 1904, about 75 mosquitoes, fed from twelve to 72 hours previously on infected animals, fed on this guinea-pig. The blood was examined regularly without result until the animal was accidentally killed on August 8, 1904; no trypanosomes or signs of trypanosomiasis were seen at the autopsy.

III. EXPERIMENT WITH *SIMULIUM*

During the rainy season there were legions of *Simulium* about the sleeping sickness hospital at Leopoldville. It was hoped to do a series of experiments with them, but they were found to be so difficult to manage that the idea was given up.

Some of these flies, freshly caught at the hospital, were, however, fed on a guinea-pig; five of them had in addition fed an hour previously on a heavily infected guinea-pig. The animal was kept under observation for 22 months; it never became infected.

IV. EXPERIMENTS WITH *ORNITHODOROS MOUBATA*

Because of the following observation, it was thought that this tick might disseminate sleeping sickness.

At Nyangwe, out of four women presenting themselves as suffering from fever after the bites of *Ornithodoros moubata*, three were infected with trypanosomiasis. All four women were wives of an arabised chief and inhabited a house, where *Ornithodoros moubata* swarmed, in a well-kept village situated in open country; tsetse-flies were consequently very rarely seen in the neighbourhood of their dwelling place.

* The larvae were used several times and therefore some of those employed in experiments 107a, 108 and 60 had possibly fed several times previously on infected animals.

INTERRUPTED FEEDING

To infect a susceptible animal with **Trypanosoma gambiense** *by the bites of Ornithodoros moubata which had fed from three to five minutes previously on a heavily infected animal.*

EXPERIMENT 1.—Guinea-pig. From August 29 to October 25, 1906, 362 ticks fed. The blood was examined almost daily without result until January 3, 1907, when the animal died of pneumonia; no trypanosomes or signs of trypanosomiasis were seen at the autopsy. A rat sub-inoculated on November 18 did not become infected.

EXPERIMENT 2.—Guinea-pig. From September 27 to November 24, 1906, 76 ticks fed. The blood was examined daily until January 21, 1907; no trypanosomes were seen: a rat sub-inoculated on December 19 has not become infected.

EXPERIMENT 3.—Rat. On November 26, 1906, 129 ticks fed. The blood was examined almost daily up to January 21, 1907. No trypanosomes were seen. A rat sub-inoculated on December 19 has not become infected

These animals (Experiments 2 and 3) were inoculated in February, 1907, with *Trypanosoma gambiense*. Both became infected and their disease ran a usual course.

Although these experiments are far from conclusive, they seem to indicate that *Trypanosoma gambiense* is probably not easily transmitted by any of the common arthropods experimented with.*

The alternative explanation of the comparative insuccess of attempts to transmit *Trypanosoma gambiense* by *Glossina palpalis* was that the experiments had not been carried out under the best conditions.

Koch (9) suggested that trypanosomes could not be transmitted by tsetse flies unless they were taken up by the fly at some particular point in their development. He supported this suggestion by the observation that he could transmit cattle trypanosomes most easily by the bites of tsetse flies previously fed on long-infected animals whose blood contained very few trypanosomes.

This suggestion of course premises some sort of development of the trypanosome in the tsetse fly. From what is known of other haematozoa there seems to be no reason why there should be none.

* In this connection the following observations should be noted. Greig and Gray (10, page 203) could not transmit the cattle trypanosomes conveyed by *Glossina palpalis* by the bites of *Stomoxys* (see page 201).

Nabarro and Greig (15) also failed to transmit these trypanosomes by *Stomoxys*.

Bruce (12, page 5), working with *Trypanosoma brucei*, found no case of spontaneous infection amongst his stock animals in an area where all sorts of biting flies, save *Glossina*, were present.

It is to be noted that these observers did not try interrupted feeding. The insects they employed, therefore, had not the best opportunity of acting as mechanical transmitters; see transmission of surra (16). Minchin, Tulloch and Gray (17) succeeded in transmitting the "Jinja" cattle trypanosome by *Stomoxys* fed interruptedly.

III. DISSECTIONS OF TSETSE FLIES

The dissections of tsetse flies commenced in the Gambia,[1] with the object of finding such a phase of the trypanosomes, were continued in the Congo.

Bruce[14] found scanty trypanosomes in the proboscis of *Glossina morsitans* fed 46 or fewer hours previously on an animal infected with nagana; it is quite conceivable that the parasites should be mechanically transmitted by the bites of such insects. It was therefore important to determine how long after feeding on an infected animal *Trypanosoma gambiense* could be found in the proboscides of *Glossina palpalis*. A series of 75 dissections* were done at periods ranging from one minute to 20 hours after feeding. It was found that red cells and trypanosomes were almost always present in the labium up to ten minutes after feeding. Later than this the parasites were frequently absent, although red cells might still be present. The longest period after feeding at which trypanosomes were found was $1\frac{3}{4}$ hours; the longest period for red cells was $7\frac{1}{2}$ hours. Red cells were seen in only six out of 25 dissections done more than two hours after feeding.

Koch[9] by pressing the bulb at the base of the tsetse's proboscis was able to obtain a clear fluid in which he frequently found trypanosomes. We found that by irritating a fly held by the wings it could frequently be made to spontaneously exude such a fluid; only once was a trypanosome seen in it, and that in a fly which had fed one and a half hours previously on a heavily-infected animal (red cells also present in fluid). It was also found that flies caught after they had fed on an infected animal frequently regurgitated in struggling a drop of blood, as large as a pin's head, which was full of parasites, many of them identical in form with those ingested. This observation was made up to 28 hours after the last feed on an infected animal.

In this connection it is well to remember that trypanosomes may live apparently unaltered in the fly's stomach for at least 48 hours. Such a regurgitation taking place during the feeding of the fly can be easily conceived as a possible means of transmission of the parasite.

* The flies previously fed and kept in test tubes were caught with forceps by the wings with as little struggling as possible. The head of the fly was cut off with a quick cut of a fine pair of scissors, in order to prevent regurgitation, and the proboscis was then detached and dissected for examination in a tiny drop of normal salt solution.

IV. MORPHOLOGY OF TRYPANOSOMES IN THE ALIMENTARY CANAL OF CERTAIN ARTHROPODA*

I. IN *GLOSSINA PALPALIS*

The observations here recorded were made on material obtained from dissections done in the Gambia and in the Congo in 1903 and the first months of 1904. Work of more immediately practical importance prevented us from giving as much time to this investigation as could have been wished, and has deferred the examination of our stained specimens and publication of our findings until now. The members of the Royal Society's Sleeping Sickness Commission and Koch have in the meantime published their work on the same subject. In many points our observations coincide with theirs. For this reason we do not give detailed descriptions of forms observed, and this especially since little can be said concerning their significance. All our work was done with *Trypanosoma gambiense* and *Glossina palpalis*. Flies were dissected and examined in fresh and stained specimens (dry films) at every period from a few minutes up to eleven days after feeding on an infected animal. Their intestinal contents, muscles, ovaries, malpighian tubules, blood and salivary glands have been examined. Only in the alimentary canal have forms been found which can certainly be connected with the trypanosomes. Active unaltered parasites were seen in the alimentary canal up to 48 hours; living, but altered, trypanosomes up to 72 hours after feeding. No recognisable trypanosomes were ever seen in the faeces of tsetse flies or in the clear fluid excreted *per anum* immediately after a fly has finished its feed.

Digestion is rapid; in a little over 24 hours after a feed the stomach is usually emptied of blood. In about 50 to 70 hours the whole intestine is empty save for a small amount of greenish-brown faeces.

After ingestion the trypanosomes become more active and apparently much more numerous; longitudinal division forms are frequent and have been seen up to 48 hours after feeding. Many half-digested parasites are distorted, vacuolated and granular, and

* Under this heading, it must be understood that controls were constantly examined.

are breaking up. They seem to be degenerating. Groups of five or more parasites agglomerated by their posterior extremities frequently occur. These usually degenerate until only shapeless débris and chromatic granules remain (on one occasion spherical forms were produced).*

In blood ingested for some days parasites approximating to the male and female types[21] become relatively much more numerous than at first. The structure of the nucleus varies. The chromatin may be collected at either pole, or in three or four large masses. It may be irregularly arranged in transverse rods or distributed in granules placed on a chromatic reticulum. Sometimes the granules are placed about the periphery of the nucleus. Chromatophilic granules occur in the cytoplasm. These have been seen in positions suggesting that they have been extruded from the nucleus.[17] Occasionally one or perhaps two large faintly pink-staining areas, somewhat diffuse, but nevertheless possessing a definite outline, have occurred in the neighbourhood of (usually posterior to) the nucleus. This area suggests the chlamydoplasm[34] observed in *Leucocytozoon danielewski*. The blepharoplast often consists of two or more granules. It is frequently very apparent that it is placed in a "clear" area and that the thickened edge of the undulating membrane ends not in it but in a pinkish basal granule or "diplosome."

Trypanosomes occur which possess a third, deeply-staining chromatic area (one-third the size of the nucleus) in addition to nucleus and blepharoplast. Other polynuclear forms resembling in appearance the polynuclear forms described by Koch† have been seen (48 hours after feeding). We can say nothing concerning their significance, but we are not prepared to follow Koch in his interpretation of them. We believe that the rounded forms, which he thinks they produce, arise from the englobation of single trypanosomes which have cast off blepharoplast and undulating membrane and become spherical. (This process has been observed in blood within three hours after its ingestion.) The bodies so formed are rounded or oval, and measure about 4·0μ to 5·0μ by 2·0μ to 3·0μ. They consist of a light-blue-stained protoplasm with definite contour and at first one, later two, a large and a small, chromatic masses. We have not seen

* Occasional confluent collections of degenerated parasites are seen which recall the "plasmodial" masses of Plimmer and Bradford (24).

† Koch gives no measurements. We cannot say whether there is any resemblance in size; our forms measure 18·0μ to 25·0μ by 3·0μ to 3·75μ.

the subsequent formation of a flagellum, and the assumption of "herpetomonas-like" characters described by Koch (their occurrence seems most probable).[34]

The spherical forms are produced in *Glossina* in the same way as in the rat flea (see below). The independent movements of flagellum and cytoplasm are most striking in parasites in the process of exflagellation. When separated the flagellum and blepharoplast quickly become motionless; the cytoplasm is mobile for a little longer. These rounded forms were not seen to divide, but from the analogy of *Trypanosoma loricatum*,[34] and from the difference in size between the smallest rounded forms (4·5μ by 3·75μ) and the average pear-shaped trypanosome (8·0μ by 5·0μ) just before exflagellation is complete, it seems possible that they may do so at this stage. After the flagellum and blepharoplast are cast off, the nucleus, at all events in some cases, fragments into from eight to ten granules. These then collect together to form a new central mass (some may remain in the cytoplasm; are any extruded?). Before the motor apparatus is thrown off from the rounded trypanosome it sometimes circumscribes the parasite so as to simulate a cyst wall—no real encystation has been observed. (Moore and Breinl[26] describe the formation of a cyst wall in similarly rounded parasites observed in trypanosome-infected animals treated by atoxyl.)

II. IN *PEDICULUS*

In July and August, 1903, a few pediculi caught on a rat very heavily infected with *Trypanosoma gambiense* were dissected and examined fresh at room temperature. Granular, indistinct, obviously degenerating parasites were of course present. The main change seen in those apparently normal was the formation of ovoid or spherical trypanosomes (5·6μ by 6·4μ to 8·32μ by 7·6μ). These had in stained specimens the same appearance as those observed in trypanosomes from the gut of tsetse flies. The following short account of one observation sufficiently describes what was seen in all:—

The alimentary tract, filled with bright red blood, was drawn from a freshly-caught louse; it was then teased out in a minute drop of normal saline. Many active normal trypanosomes were present. All were long and large at first; no stumpy forms were seen. Some of the parasites were quite granular. One hour later the normal parasites were less numerous, and pear-shaped trypanosomes were present which had lost undulating membrane and flagellum. Similar forms existed to which the flagellum was still attached by the blepharoplast. In these forms

flagellum and cytoplasm move quite independently of one another. The movements of the cytoplasm were both amoeboid and lashing. These forms, as well as some very stumpy and granular, fairly round trypanosomes, may roll themselves up into a ball. (If the flagellum be present it is usually external.) Some seemed to be enclosed in some sort of a membrane, since, although motion was violent, they never changed their spherical shape. Others straightened out for a moment to immediately curl up again. In stained specimens no definite cyst wall was ever seen to enclose a spherical parasite.

One apparently normal trypanosome was first seen at this time *inside* a large clear vacuolated cell of the stomach wall.* During the three and a half hours it was watched it remained active and unable to leave the cell. After four hours bacteria commenced to multiply in the specimen, and it was necessary soon after to discard it. No further change was observed in the spherical forms produced by the rounding of the above described "pear-shaped" parasites.

III. IN *STOMOXYS*

In the Gambia, trypanosomes, identical with those ingested, were found unchanged in the gut of *Stomoxys* up to 20 hours after they had fed heavily on a horse infected with *Trypanosoma gambiense.* Longitudinal divisional forms were seen. In one instance phagocytosis (by a cell from the horse's blood ?) of a trypanosome was seen at this period.

An interesting observation was made in a fly fed 18 hours previously. Two trypanosomes were attached to one another; the separate outlines of each parasite could be distinguished perfectly. In about an hour and a half, the two parasites had fused to form a single trypanosome. The newly-formed parasite was thicker than is normal, had a shorter flagellum and a collection of very minute refractive granules at either end and no perceptible nucleus in the usual position. After twelve hours the parasite was motionless; no further change had occurred. This phenomenon was twice seen.

IV. IN LARVA OF *AUCHMEROMYIA LUTEOLA*

The contents of the intestinal canal, the salivary glands and blood of larvae fed from six to thirty-four hours previously on animals infected with *Trypanosoma gambiense* were examined. The changes observed were very similar to those seen in the other arthropods. Living trypanosomes were seen in the fresh blood of the diverticulum up to twelve hours after feeding, and no parasites were seen in the old black, half-digested blood. At ten hours many parasites were dead; numerous agglomerated and degenerating trypanosomes were present; some of the stumpier parasites were becoming rounded. The intestinal contents contained many bacteria; perhaps for this reason comparatively few small spherical forms of the trypanosomes were seen.

* Battaglia (quoted from 19, page 113) reports having seen intracellular forms of a bat trypanosome and of *T. lewisi* (?).

V. IN ANOPHELINES*

A large series of Anophelines (*Pyretophorus costalis*) was dissected at periods ranging from twelve hours to eleven days after feeding on experimental animals heavily infected with *Trypanosoma gambiense*. In the interval between feeding and being dissected, the mosquitoes were fed every two days on uninfected animals. Alimentary canal, malpighian tubules, ovaries, and salivary glands were carefully examined, and forms recognisable as connected with the trypanosomes were seen only in the alimentary canal; at first some confusion was caused by the presence of the sporozoites of a coccidium-like protozoon.[34] After 42 hours no trypanosomes were seen. From 12 up to 42 hours after feeding the changes similar to those described in the gut of other arthropods were observed. Nothing peculiar was seen.

V. DISCUSSION

When malarial parasites are ingested by a receptive anopheles, only those parasites survive which are prepared for development in the environment provided by their new host. The remainder die. If the blood is heavily infected there is therefore necessarily a large number of degenerating parasites present in the insect's alimentary canal. The same may be true of trypanosomes ingested by tsetse flies. The mass of degenerating parasites confuses one's perception and makes it extremely difficult to pick out the forms capable of further development.

It is by examining in fresh coverslips and, later, stained preparations, the parasites in trypanosome-infected blood taken from the gut of insects, that the changes invariably taking place under such conditions can best be determined. It seems reasonable to deduce that forms invariably appearing are those best suited for the new environment, and that they may be possibly concerned with the further development of the parasite. Such forms should therefore be most carefully

* A paper (33) important in this connection was received while the present communication was in the printers' hands. Its authors show that *Trypanosoma brucei* survives ingestion by mosquitoes for only 36 hours; it remains virulent to mice for only 14 hours. The morphology of the ingested parasites is not reported on.

watched. Up to a certain point observations on trypanosomes in cultures may be very valuable here.

Bouet (Culture du Trypanosoma de la grenouille, Annales de l'Inst. Pasteur, Vol. XX, No. 7, page 364) has obtained a very large part of the "swarm" cycle (34) of this parasite in cultures.

Novy and MacNeal, in their studies on mammalian and bird trypanosomes, illustrate forms which are interesting in this connection. Life History of *Trypanosoma lewisi* and *Trypanosoma brucei*, Journal of Infect. Diseases, Vol. I, No. 4, Nov. 5, 1904, p. 517. On the trypanosomes of birds, Journ. Infect. Diseases, Vol. II, No. 2, March 1, 1905, p. 256.

Cerquira, in a thesis published at Rio de Janeiro, describes a most interesting development in bird trypanosomes in cultures. It bears evident analogies to the multiplication process described in *T. loricatum* (34) (reviewed in Journal of Tropical Medicine, April 1, 1907, p. 118).

A process which has been seen by many observers in trypanosomes ingested by arthropoda is the formation of spherical bodies without undulating membrane, flagellum or, at first, differentiated blepharoplast. Similar forms have been reported by many different authors in most of the mammalian trypanosomes when they are placed in various "unfavourable circumstances,"[19] such as in moribund animals, in the cerebro-spinal fluid, in organs, kept preparations of fresh blood, cultures, gut of lice, fleas and biting flies.

These rounded parasites bear a most striking resemblance to the rounded form of *Trypanosoma loricatum* which commences the interesting cycle of "swarm" development first described by Danielewsky.[27] These forms of the mammalian parasites are formed in much the same way as are the rounded frog trypanosomes. Does their further development proceed in the same way? (see footnote to page 62 of [27]). The work of the following observers shows that there may be some resemblance between their subsequent stages.

Bradford and Plimmer (24) describe the rapid division of spherical "amoeboid forms" of *T. brucei* in the blood of a vertebrate host.

Prowazek (22) also reports the division of such bodies in *T. brucei*; and describes the formation of similar spherical bodies in *T. lewisi* ingested by a louse; and he suggests that these later develop a flagellum.

Koch (9) describes the formation of rounded forms, which subsequently develop a flagellum, in cattle trypanosomes ingested by *Glossina*. In their development these parasites pass through a herpetomonas-like stage.

Lingard (28) describes certain developmental processes in trypanosomes of Indian cattle where similar rounded forms occur.

Moore and Breinl (26) have observed similar forms in *T. gambiense*, especially in animals treated by atoxyl; their further development has not been followed. A second type of rounded form, produced by the extrusion of the nucleus from an ordinary trypanosome, was observed to produce a minute flagellum.

Durrant (29) and Holmes (30) describe similar rounded forms in *T. evansi*. The latter described them as undergoing development. He also describes the second type of rounded trypanosome and its development as do Breinl and Moore.

It is impossible to say how far this resemblance extends. At all events, the above observations prove that the spherical forms represent a stage in a definite cycle of development other than that of longitudinal division, the only definitely developmental phenomenon previously established in the life history of trypanosomes. The observations of Holmes, Moore and Breinl show that there is possibly a third line of development in mammalian trypanosomes, in addition to the multiple segmentation first described in *Trypanosoma lewisi*.[32]

Some of the rounded forms of mammalian trypanosomes doubtless degenerate and die, as do similar forms of *Trypanosoma loricatum*. But this constitutes no reason for calling all rounded forms degenerative or involutive, as has been so frequently done. The greatest caution must be used in applying such terms; it is chastening to remember that only ten years ago a flagellating malaria parasite was widely believed to be a degenerating form!

Conjugation* has not been seen certainly. Possibly it does not occur during multiplication in *Trypanosoma loricatum* by the "swarm" cycle; but it is not at all impossible that a union of sexually differentiated individuals may occur at some other period.†

It has been stated that the occurrence of a cycle (probably sexual) in mammalian trypanosomes ingested by tsetse flies is inherently improbable. The following are the points most usually cited to support this view:—

(1) It is said that there is no need for a sexual cycle since trypanosomes may be transmitted for an indefinite period mechanically, from animal to animal by simple inoculation and by fly bites, or

(2) that the trypanosomes may be kept for very long periods in cultures.

* The word is used in its wider sense.

† Moore and Breinl (26) describe nuclear phenomena in *T. gambiense* which, it is suggested, may be explained as a sexual act. Such a self-fertilisation may perfectly well occur in a parasite in addition to the conjugation or copulation of sexually differentiated individuals.

(3) That another adequate method of reproduction (now probably at least two) already exists.

(4) That the contents of the intestinal canal of tsetse flies previously fed on trypanosome-infected animals when inoculated into susceptible animals does not produce trypanosome infection (nor, in the cases of bird trypanosomes and mosquitoes, will they produce cultures of trypanosomes).

None of these arguments is valid. Since those who uphold them usually oppose Schaudinn's work[21] on *Haemoproteus*, etc., we shall not mention it in answering their arguments, but shall refer only to the work of other authors. Each objection is answered separately by referring to some occurrence contrary to it in the known life history of some other protozoon.

(1) Human malaria can be transmitted by simple inoculation, *Trypanosoma lewisi*, *Piroplasma bovis*[31]* and *Haemamoeba relicta* can be propagated indefinitely by inoculation. In all four of these protozoa a definite sexual process occurs when the mature parasites come into a favourable environment, the alimentary canal of the required arthropod.

(2) *Trypanosoma lewisi* has been successfully cultivated. Prowazek[22] describes a sexual conjugation which it undergoes in lice.

Although no other pathogenic protozoa have been cultivated with the same success as the trypanosomata, it is well known that certain non-pathogenic protozoa may be kept for very long periods in pure cultures without the appearance of any sexual forms. On the production of circumstances impelling them, sexual forms may appear in these cultures. Granting that the sexual forms are absent from, not merely unidentified in, the usual cultures of trypanosomes, it by no means follows that they may not be produced under other circumstances. The conditions causing their appearance are not present in the ordinary culture medium. For the same lack of "impelling causes" the periodic absence of infective sexual forms from the blood of infected animals is easily conceivable. The question naturally arises, what are the conditions "impelling" the production of sexual

* Professor Nuttall permits us to state that he is of opinion that *Piroplasma canis* may be transmitted indefinitely from dog to dog by inoculation. He has already transmitted the disease in this way for more than twenty generations.

forms of trypanosomes? The analogy of malaria in man and of protozoa in cultures suggests an answer—chronic infections.*

(3) Young malarial parasites (ring forms) divide by direct division; a cycle of sporulation exists, yet there is also a sexual cycle.

(4) These facts need only signify that the parasites were not, at the moment of inoculation, in a state favourable to cultivation or to the production of infection in a vertebrate host.

Coccidium schubergi is parasitic in a centipede. Infection takes place by the digestive canal. If an infected centipede be eaten by another, infection may be conveyed by almost every stage of the parasite ingested. The immature gametocytes, however, are unable to infect; although they are in their normal host their surroundings are unsuited to their further development. They consequently die.

In cover-slip preparations the malarial gametocyte is fertilized, development goes but little further; the surroundings are not favourable. Why should the sexual cycle of a trypanosome necessarily occur in the same medium as its asexual one? Crescents, males at least, adult malarial parasites, if left in man degenerate; the young parasites die in the mosquito. The surroundings suited to one form are not to the other. Why should the hypothetical sexual form of a trypanosome develop equally well in either invertebrate or vertebrate host?

The cited arguments proffered against the existence of a sexual cycle of trypanosomes in tsetse flies are contradicted by these known facts. What are the arguments in favour of it? They are briefly:—

(1) The analogy of other protozoa.

(2) The occurrence of parasites possessing sexual characteristics in the blood of infected animals† (compare *Trypanosoma dimorphon*).

(3) Prowazek's observations on the development of *Trypanosoma lewisi* in the rat.[22]

(4) Koch's incomplete observations on the development of cattle trypanosomes in *Glossina*.[9]

(5) Supposedly sexual forms, somewhat similar to some of those

* Koch (9) states that his *Glossinae* only became infected and the trypanosomes they ingested only developed if their feed had been made on a long-infected animal in whose blood the parasites were scanty.

† The acknowledgment of the nature of these forms of course depends in some measure on a recognition of at least a part of Schaudinn's work (21). They have been reported in many trypanosomes by many independent observers.

described by Koch, are reported in *Trypanosoma gambiense* ingested by *Glossina palpalis*.*[17]

(6) We have observed similar forms to (5) and in one instance at least the fusion, we believe, of two living *Trypanosoma gambiense* in blood from the gut of a *Stomoxys*.

The arguments on neither side are conclusive, but we believe those to be far the stronger which support the existence of a sexual cycle in the life history of the mammalian trypanosomes, possibly in the tsetse fly.

It is, we believe, by the most patient and long-continued observation of living, individual parasites in trypanosome-containing blood (probably freshly ingested) from the gut of tsetse flies that a definite answer can best be given to the whole question. The work should be done under natural conditions in Africa and, naturally, the observations on living parasites must be controlled and supplemented by the study of stained specimens. The observer must approach the subject in a receptive mood and freed from preconceptions.

VI. CONCLUSIONS

From the consideration of all the facts we state the following propositions:—

It is known that,

(1) Mammalian trypanosomes may be mechanically transmitted by the bites of blood-sucking arthropoda.

(2) A cycle of development of the trypanosomes exists in which occur spherical parasites formed by the throwing off of blepharoplast, undulating membrane and flagellum. These forms exist in both the invertebrate and the vertebrate hosts; conjugation probably plays no part in the production of this form. A possibly distinct cycle is

*These authors also describe "fly trypanosomes" occurring in the gut of tsetse flies which, in common with Novy (23), they consider to be entirely distinct from the mammalian trypanosomes. We have not seen trypanosomes, recognisable as other than those ingested, in the flies examined by us. In one instance, we found trypanosomes in the gut of a freshly-caught *Glossina palpalis*, but fresh mammalian blood was also present; another parasitic protozoon, not a flagellate, was also present. These fly trypanosomes (20) have an encysted stage which leads Minchin to suggest that their cysts may be excreted, ingested by a vertebrate host, and so take part in a "contaminative cycle." The hypothesis is a most interesting one. Minchin states that the sexual forms of *T. gambiense* observed in the gut of tsetse flies examined disappeared and after 96 hours parasites could not be found. Nothing "resembling a trypanosome" was found in the various parts of the fly examined later. Why should developmental forms or the product of the *copula* of trypanosomes be trypanosome-like in shape?

represented by the spherical parasites, consisting almost entirely of nucleus, which result from the disintegration of trypanosomes[26, 30].

(3) There is reason to suspect that a sexual cycle may also occur; it may be in either or in both hosts.

(4) The rapid spread of sleeping sickness cannot be fully explained by (1) alone; the cycles of development mentioned in (2) and (3) probably play a very important part in the transmission of the disease.

RUNCORN RESEARCH LABORATORIES
April, 1907

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CATTLE TRYPANOSOMIASIS IN THE CONGO FREE STATE

CATTLE TRYPANOSOMIASIS IN THE CONGO FREE STATE*

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I. INTRODUCTION

When Europeans first came to the Congo there were practically no cattle in the greater part of the central area of what is now the Free State. Since their advent cattle have constantly been imported from neighbouring African colonies.†

They have been sent to many posts in different parts of the Congo with varying success. As a rule, it is said that cattle tend to do best in plain country where there are no buffaloes.

* Much of the information contained in this paper was communicated in 1904 to the Government of the Congo Free State in unpublished reports.

† The natives in the highlands about Lake Kivu possess large herds of cattle. As a rule the Kivu cattle (Fig. 3) are rather small and have long, upstanding, slightly-curved horns (*Bos aegyptiacus* ?). Some of them are of the humped zebu type. The cattle in the parts of the Congo visited by the expedition[1] generally approximate to the European type. Tsetse flies are said not to be present in the Kivu district. Game is plentiful. The mortality among cattle taken from Lake Kivu to other districts is very great. From herds of 20, only two or three have remained on arriving at Kasongo! It was found that more cattle died in herds which travelled only every third day than in those driven hard. The cause of the mortality is therefore probably not fatigue and lack of food, but disease. Seventy-five head of cattle taken North from Rutshuru did well until the forest was entered. They then commenced to die, and at Mawambi all were dead.

II. EXAMINATION OF CATTLE

We examined the cattle of only a few of the more important herds; those existing at the remaining places visited are only briefly considered, while herds unvisited are not mentioned.

At ZAMBIE there is a herd of about 190 cattle, all in excellent condition; in 1904 there were 52 births, and no deaths from disease. Those in charge recognise no disease resembling trypanosomiasis.

At the neighbouring island of MATEBA there is a herd of about 5,000 (?) cattle; said to be in perfect condition. The original animals at both Zambie and Mateba were imported from Portuguese West Africa (San Paul de Loanda, etc.), Walfish Bay, the Canary Islands, and occasionally from Europe. Both places are on low-lying grass plains, and tsetse flies, although probably present, were not seen (October, 1903), and big game was practically absent. Living animals are regularly sent from St. Paul de Loanda, from Mateba and from posts in the Lower Congo to Leopoldville for food; two said to come from Mateba (?) were found to be infected with trypanosomes on their arrival at Leopoldville.

The LEOPOLDVILLE herd, kept only for food purposes, is heavily infected [2, 3].

At YUMBIE cattle have been present since 1896, but they have died as fast as they reproduced and the herd has not increased. The principal cause of death is "colic" (?). This disease is probably trypanosomiasis (see below). When we saw the herd in July, 1904, the twelve animals present, with one or two exceptions, seemed in fair condition. In June, 1905, the ten animals then composing the herd were seen by Broden.[4] They were all in wretched condition and three of them had trypanosomes in the peripheral blood. In September, 1904, six heifers arrived from Dolo (near Leopoldville). From October to June, 1905, there were eight deaths; all probably from trypanosomiasis.

Yumbie is placed on a high grassy plain with neighbouring forest. *Glossina palpalis* and *fusca* (?) are present. Large game is plentiful.

Six young cattle were brought to BASOKO from Nouvelle Anvers in 1898. The herd now numbers eight (only two deaths from accidents have occurred); they all seem to be in excellent condition. The herd grazes in cultivated ground surrounded by thick forest. *Glossina palpalis* present, but rare; big game not plentiful.

LISENGI. No deaths recorded; cattle present for six years. They graze in uncultivated coffee plantations surrounded by forest. No buffaloes, many antelopes present. *Glossina palpalis* not seen, probably present.

At KIRUNDU there were a few miserable-looking trypanosome-infected cattle, in the hands of the natives.

EALA, August, 1904. Before reaching Eala a report was received from the Veterinary Surgeon resident there, M. Bertolotti, in which he described an obscure disease affecting the cattle under his care. The symptoms resembled those of trypanosomiasis, and on examination we succeeded in finding the parasites in one cow (Fig. 1) in which the disease was well advanced (no trypanosomes were found by a single centrifugalisation of the blood of five other animals chosen from the herd for their lack of condition). The extent of the disease at Eala is shown by the following review of the cattle records.

There is at present a herd of 43 cattle at Eala. These represent the remnants and product of 51 cattle sent here from the Lower Congo in May, 1901, one bull and ten cows (of these only two cows are still alive), and from April to May in 1902, five bulls and thirty-five cows (of these one bull and 26 cows are still alive).

The great mortality amongst these animals has been almost exclusively due to trypanosomiasis.* Calves born at Eala (and at Nouvelle Anvers) have suffered specially from this disease; of 36 born at Eala only 14 are living (for example, of 15 bull calves born between August, 1902, and January, 1904, 12 were dead in August, 1904, of trypanosomiasis). Some of the young animals born here which are still alive are undersized and obviously ill-developed.

Histories of the Infected Animals

Cow No. 10.—Brought to Eala from the Lower Congo in 1902. July 23, 1904: Is at present extremely thin and scarcely able to move (Fig. 1). Superficial glands are enlarged (M. Bertolotti regards this as one of the most constant symptoms of the disease; diarrhoea is also a usual symptom). Blood centrifugalised; no trypanosomes seen. August 5, 1904: Animal still thinner; hardly able to stand because of weakness; but still has appetite and munches grass. No oedemas or petechial haemorrhages observed. Blood centrifugalised and prescapular gland punctured; trypanosomes found by both methods. The animal was killed by injecting air into jugular vein and an autopsy was made at once.

* Clinical diagnosis; the parasites were not observed in affected animals until July, 1904.

Autopsy.—Animal is very thin ; muscles pale. Organs all normal, spleen not enlarged. Superficial lymphatic glands twice as large as normal, watery, not congested. Deep glands all much enlarged, about half of them (especially in abdomen) much congested—almost haemorrhagic. Examination for trypanosomes : Trypanosomes were found in the blood (20 to a coverslip preparation) only after centrifugalisation (five coverslip preparations of uncentrifugalised blood were examined without result). They were also seen (three to a coverslip) in a preparation of gland juice obtained from the prescapular gland by a hypodermic syringe before the animal was dead; none were seen, however, in nine preparations of the juice of glands from various parts of the body made within one to two hours of the animal's death. Many parasites were found in the deposit obtained by centrifugalising slightly turbid fluid from the pleural (60 c.cm. present), the peritoneal (500 c.cm.) and the pericardial (35 c.cm.) cavities. It was noted that the trypanosomes seen in these fluids were almost invariably attached to a white cell. Forty-five c.cm. of cerebro-spinal fluid taken immediately after death was centrifugalised and most carefully examined. It contained no trypanosomes and no red or white cells, but it was made slightly opalescent by a peculiar flocculent material present in small quantities.



FIG. 1.—COW, No. 10, at Eala. August 23, 1904

Inoculations.—Two rats (Exp. 137, 138) each received 4 c.cm. of blood from this cow intraperitoneally on July 23. Neither had become infected* on August

* The blood of all the experimental animals mentioned in this paper was examined, almost daily, in three-quarter inch square fresh coverslip preparations until either the animal became infected or the experiment was discontinued.

3rd, so they were then both re-inoculated intraperitoneally, one with 5 c.cm. and the other with 3 c.cm. of a mixture of gland juice, from the same cow, and sodium citrate solution showing one trypanosome to five fields.

A guinea-pig (Exp. 142) was at the same time inoculated intraperitoneally with 4 c.cm. of the same fluid. All three animals were carefully examined; none ever became infected.

The herd at Eala grazes on low-lying land surrounded by forest. *Glossina palpalis*[1] exists in small numbers (July, 1904). Big game is scarce.

NOUVELLE ANVERS, August 15th, 1904. At Nouvelle Anvers five of the thinnest of the 34 cattle present were examined; two had trypanosomiasis. The first cattle were brought here in 1896. From then to 1900, 16 head (thirteen cows, three bulls) were imported from the Lower Congo (two from the Canary Islands). Since 1898, 48 calves have been born; ten have died (one from the results of abortion, nine from fever and unrecognised diseases), and several have been sent to other posts. Of the original animals twelve are dead (one slaughtered, 11 of insufficiently described diseases).*

It is most important to note that seven of these animals came from Stanley Pool (Kinchassa), and all the animals sent from Kinchassa to Nouvelle Anvers have died in from one to four years after their arrival. At present there is only one animal (Bull 18, arrived March, 1904), from Stanley Pool (Leopoldville) and it has trypanosomiasis; Dr. Broden has shown trypanosomiasis to be common among cattle near Leopoldville.

Though it cannot definitely be said that the animals from Stanley Pool did not contract trypanosomiasis at Nouvelle Anvers, it is quite possible that they may have been infected with trypanosomes before their arrival there. If this is the case the herd has contained infected animals since 1899 at least. Its losses since then have certainly been heavy, but the percentage of mortality is smaller and the disease seems less virulent than at Eala. For instance, one cow born at Nouvelle Anvers in 1896 and another brought there from the Canaries in the same year are still alive and well

STEER 17.—Born here December, 1902. In fair condition; supposed to be healthy; lymphatic glands slightly enlarged.† Fairly numerous trypanosomes seen in blood and gland juice.

*When the cause of death is ill-defined, it is usually because the animal has gradually wasted and died. Such cases are usually ones of trypanosomiasis.

†One of the animals in which a most careful examination failed to reveal trypanosomes also had slightly enlarged glands.

BULL 18.—Age, two (?) years. Imported from Leopoldville, March, 1904. In good condition; supposed healthy; lymphatic glands slightly enlarged. Fairly numerous trypanosomes seen in blood and prescapular gland juice.

Inoculations.—Rat (Exp. 145) inoculated Aug. 15 intraperitoneally with 2 c.cm. of blood from this animal; did not become infected.

Monkey (*Cercopithecus schmidti*) (Exp. 143) received 8 c.cm. of blood intraperitoneally, August 15, 1904. Trypanosomes appeared in its blood September 22. They soon disappeared, and were only occasionally seen during 1904. During 1905 the monkey seemed quite well, and trypanosomes were not seen in its blood, which was examined at intervals. On January 1, 1906, it was very ill, weak, dull and apathetic. A single degenerated (?) trypanosome was seen in two coverslip preparations. Animal died next day; immediate cause of death was miliary tuberculosis.

A rabbit (Exp. 144) received 3 c.cm. of blood intraperitoneally; never became infected; under observation for six months.

The herd at Nouvelle Anvers grazes for the most part on cultivated land. Big game is not very abundant, but biting flies of several sorts (*Glossina palpalis*, *Stomoxys* and *tabanids*) are present.

ROMEE, September 12th, 1904. One or two animals now present have been here since 1897, but the great majority of the cattle[19] have been brought here since the end of 1902. Most of them are of Lake Kivu stock and came here by way of Nyangwe. A few head came from the Nile. Only nine deaths have occurred since March, 1903; all were cattle which had arrived in poor condition two or three months previously. It therefore appears that this herd does not lose so many animals from disease (trypanosomiasis ?) as do those at Eala and Nouvelle Anvers. For example none of the calves born here (first in 1899) have died. We were shown ten animals that had been kept apart from the herd of 25 head because of their lack of condition. Six of them (arrived at Romee, July, 1904) were examined.* Four had trypanosomiasis; three of these were decidedly thin and had enlarged lymphatic glands, the fourth had no big glands and was comparatively well-fleshed. All had temperatures of over 101·2°F. All four infected were of Kivu stock; two had come from Stanley Falls and the other two were from Nyangwe.

Three rats (Exp. 138a, 149, 151) inoculated intraperitoneally with blood from Cow No. 2 became infected in three, six and eight days and died in 18, 23 and 21 days.

* In July, 1905, one of these animals was still living and seemed perfectly well; the other three had been slaughtered.

Two guinea-pigs (Exp. 148, 152) inoculated in the same way, one from one of the above rats, the other from the cow, showed parasites in twelve and fifteen days and died in 18 and 23 days of ruptured spleens (much enlarged and friable), in one case after slight violence.

A rabbit (Exp. 147), weight 5 lb., became infected in nine days. Parasites were at first numerous; they then became scanty and were only seen from time to time until January, 1905. None were seen after this. Death from pneumonia October 21, 1905. Spleen not enlarged, firm (5 × ·8 × ·6 cm.), capsule thickened. None of glands much enlarged. Animal very fat.

Monkey (*Cercopithecus* ?) (Exp. 146), weight 5½ lb. Inoculated September 13, 1904, with blood from one of the rats; showed trypanosomes in its blood three and six days later and never again. Death from pneumonia December 16, 1905. Spleen slightly enlarged (5 × 3 × 1·5 cm.), firm; glands not enlarged; animal fat.

The herd grazes on high, wooded, cultivated grounds. Small antelopes are not uncommon. *Glossina palpalis* is rare.

LOKANDU, October 21st, 1904. Sixteen head of cattle present. In January, 1902, there were 23; since then 13 have been sent away (nine to Romee) and three have died. In only two of the deaths were symptoms resembling trypanosomiasis present.

We examined eight heads; five had trypanosomiasis. Three, all infected, of those examined were calves born here. All were thin, had dry noses, much-enlarged glands (to 3 by 4 by 10 cm.) and slight temperatures (101·5°F.).

In two of them there were distinct patches of oedema in the loose skin of the throat. Five of those examined were adult cows imported from Nyangwe or Kasongo. All were in fair condition; only one had enlarged glands. This one, and one other in whom there was no glandular enlargement, had trypanosomiasis. The one with enlarged glands had a temperature of 106·8°F. Trypanosomes were seen in small numbers in the blood of only three of these five infected animals. They were seen in much larger numbers in the gland juice in each case.

Inoculations.—A guinea-pig (Exp. 153) was inoculated intraperitoneally on October 22, 1904, with 7 c.cm. of blood, from one of the calves, containing three trypanosomes to a coverslip preparation. It was examined carefully until January, 1905, but trypanosomes were never seen.

These cattle had scanty pasture in coffee plantations surrounded by forest. Antelopes and *Glossina palpalis* not uncommon.

SENDWE, November 5th, 1904. Four head of cattle were sent here from Kasongo in 1900. The two bulls imported died soon after their arrival, so the remaining animals were sent away to Matampas.

In May, 1904, eight head passed through on their way to Romee. Two died here; only four reached Romee alive (we examined none of them for trypanosomes). In May, 1904, ten heifers on their way to Romee arrived here by Kasongo from Lake Kivu. They were not in good condition; but this was at the time explained by a two months' stay at Kibombo—poor pasture. A month after arrival they commenced to die, and in three months five were dead. All had the same symptoms, "gradual wasting and loss of vigour." The remaining animals seemed in fair condition, but all of the four examined had enlarged glands (see Fig. 2) and temperatures (to 104·4°F.). Three of them had trypanosomiasis. Some of them were later sent to Lokandu.



FIG. 2.—CALF at Sendwe; to show enlarged gland.

The pasture at Sendwe is poor; *Glossina palpalis* and big game are plentiful.

NYANGWE, November 14th, 1904. A herd, now numbering about 60 head, has been here since the end of the Arab war (1894). It is

largely made up of captured cattle, the majority of which probably came from the neighbourhood of Lake Kivu. The cattle are at present in splendid condition. Since 1900, 45 calves have been born. During the same period there have been only five deaths, none of them from any disease resembling trypanosomiasis.

The country surrounding Nyangwe is high, grassy plain. The herd never grazes further than one and a half miles from the post and never goes near a swampy area towards the South. The country about Nyangwe is thickly populated and extensively cultivated. There is, therefore, little large game in the neighbourhood. Tsetse flies were not seen anywhere about the post nor on the plains where the cattle graze. They were seen, however, along the river bank a mile and a half further South.

The blood of 20 cattle chosen because they were less plump than the remainder of the herd was examined; gland puncture was done in 17 in which the glands were sufficiently large. Trypanosomes were not found.

KASONGO, November 25th, 1904 April 27th, 1905. There is a herd here of about 260 cattle. Many of them were collected by the Arabs; additions have been made by the whites. The animals reproduce well and the herd has been largely increased by local births; deaths from diseases are very rare. Its original members were probably largely from Lake Kivu. Some probably came from the Eastern and Southern shores of Lake Tanganyika. Very few, if any, were ever brought directly from Katanga, far to the South, or from Lusambo, in the West.

One hundred of the cattle were examined for trypanosomiasis during our stay at Kasongo, five were found to be infected. All five were found in the first twenty-two examined. This is noteworthy, since the thinnest animals were taken first. All the animals were examined in the same way and each was gone over but once. Preparations of gland juice and blood were examined and the temperature was taken in each case.

The infected animals were kept under observation for several months. The rareness with which parasites were seen in them during this period (see charts) strongly indicates that many cases remained undetected by our examinations, and therefore that the proportion of cattle infected is much more than five per cent.

ZILANI No. 8 (Nov. 15, 1904).—A young cow, born at Kasongo. She is rather thin and the natives say she is ill; but there is no oedema, staring of the coat, &c. Largest prescapular gland 12 × 5 cm., largest femoral gland 9 × 3 cm. Trypanosomes were found in gland juice, not in blood (see Chart I). Inoculations (see below, page 243).

MOIUMA No. 33 (Nov. 25, 1904).—A young cow, born at Kasongo. Apparently quite healthy. Prescapular gland 12 × 5 cm., soft; femoral 6 × 3 cm. Trypanosomes present in blood and gland juice. The course of the disease is given by Chart II up to February 8. From then to April 19 (observations ceased) the temperature varied, without any sudden changes, between 101° F. and 104° F. Blood and gland juice were examined at intervals; parasites were seen during this period by gland puncture, only on March 16.

Inoculations.—Monkey (*Cercopithecus schmidti*) (Exp. 175), January 11, 1905. Inoculated intraperitoneally with 18 c.cm. blood showing one trypanosome to cover; under observation until February 8 (when inoculated with spirochaetes); never infected.

KEOWKOSIKO No. 60 (Nov. 25, 1904).—Cow, born here. It is in fair condition, perhaps slightly thin. Prescapular gland measures 10 × 5 cm., femoral 8 × 2 cm.; both are soft. Chart III shows the course of the disease up to February 17. From then to March 9 the temperature continued in the same way and parasites were only seen on March 8, 9, 10. The temperature became irregular and rose constantly until 105° F. was touched on March 22 (no trypanosomes in blood). The temperature then became lower and no parasites were seen in blood or gland juice up to April 18, when they were in the blood in small numbers. Observations ceased.

LIBOKO No. 29 (Nov. 30, 1904).—Fair condition; glands slightly enlarged. Trypanosomes present in gland juice, absent from the blood. As is seen by Chart IV, the parasites were never again seen in this animal.

YOLO No. 9 (Dec. 1, 1904, Fig. 3).—A steer, adult, born at Nyangwe, has been here for three years. Prescapular gland is 15 × 5 cm.* and soft; femoral is 10 × 2 cm., soft and moveable. Trypanosomes were seen in gland juice and not in the blood. The course of the disease up to April 14, when the animal was killed for the autopsy, is indicated on Chart V.

Autopsy.—The animal was killed by injecting air into the jugular vein. The examination was commenced immediately after death. No subcutaneous fat or oedemas; muscles dark. Abdomen contained 400 c.cm. clear fluid; mesenteric vessels very turgid, omentum and retroperitoneal tissues oedematous. Thorax: pleurae contained no fluid, pericardium 300 c.cm. Between diaphragm and base of pericardium there was a layer of oedematous tissue 1 cm. in thickness. The fat about base of heart and along its vessels was brownish-canary-yellow in colour and was very oedematous, as was also the connective tissue of the posterior mediastinum. None of the organs showed any striking macroscopic change (spleen weight 1½ lb., substance firm). Marrow of long bones yellow. *Lymphatic glands:* Just before death the prescapular gland was excised. It was surrounded by a capsule of oedematous connective tissue ·75 cm. thick; it measured 11 × 6 cm. All the glands were enlarged and all were firm but distinctly "watery," so that the fluid followed the knife. The larger glands in all parts of the body were pale with darker cortex. Some of the smaller ones (1·5 × 1 cm.) along the small intestine, at the base of the mesentery (2 × 3 cm.) and about the bronchi and trachea (1·5 × 1 cm.) were congested or haemorrhagic. Immediately after death lumbar

* The measurements given for these glands are of course estimates, and include the capsule. This gland when excised on April 14, 1905, actually measured 11 × 6 cm.

puncture was done at about the sixth vertebra with a four-inch needle. About 4c.cm. of clear fluid was obtained. Fresh preparations of the following organ juices and body fluids were examined, juice from the prescapular, mesenteric and tracheal glands, liver, spleen, marrow from long and short bones, blood, pericardial and cerebro-spinal fluid. Parasites were only seen in the gland preparations.

Inoculations.—Dog* (Exp. 227), April 13, 1905. Inoculated intraperitoneally with about 150 c.cm. of defibrinated blood from the jugular vein. Died December 12, 1905; no autopsy. No trypanosomes seen.

Dog (Exp. 229), April 13, 1905. Inoculated as above with 92 c.cm. of blood. No trypanosomes seen until December 1, 1905, when killed because of mange.

Dog (Exp. 229), April 13, 1905. Inoculated as above with 150 c.cm. blood. Killed because of mange, December 1, 1905. Trypanosomes were never seen in the blood.

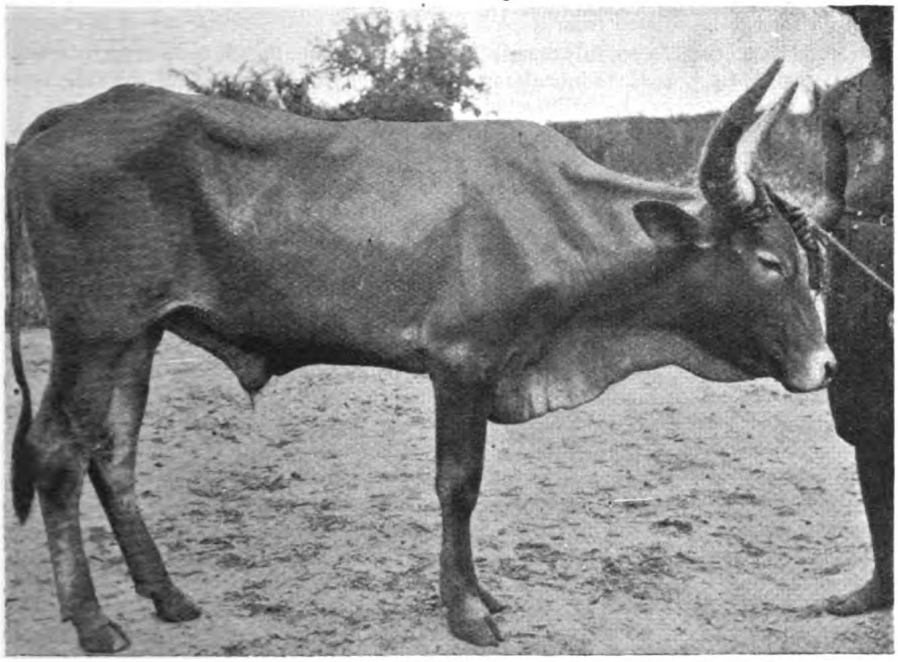


FIG. 3.—STEER, *Yolo*, at Kasongo. Note emaciation. The type of horns and tendency to hump indicate a Lake Kivu origin.

Experimental Inoculations at Kasongo

Most of the animals inoculated at Kasongo were so frequently re-inoculated that it has seemed best to treat the majority of them under a separate heading.

* The dogs used in all these experiments were native ones bought at Kasongo.

Rat (EXPERIMENT 132), July 6, 1904. Inoculated with buffalo blood (*Bos nanus*); never infected. (Exp. 221) April 3, 1905. Inoculated with 5 c.cm. blood from *Yolo* showing four trypanosomes to cover. Died August 20, 1905 (no autopsy). Never infected.

Rat (EXPERIMENT 133), July 6, 1904. Inoculated with buffalo blood; never infected. November 8, 1904, re-inoculated (8 c.cm.) with buffalo blood. (Exp. 222), April 3, 1905, re-inoculated with 5 c.cm. blood from *Yolo* showing four trypanosomes to cover. September 8, cataract in one eye, blood in anterior chamber of the other. December 8, TRYPANOSOMES FIRST SEEN IN BLOOD. December 15, died, blood swarming with trypanosomes. Glands and spleen enlarged, organs otherwise healthy.

Rat (EXPERIMENT 134), July 6, 1904. Inoculated with buffalo blood; never infected. (Exp. 177) January 11, 1905, re-inoculated with 12 c.cm. from *Moiuma* showing one trypanosome to a cover; never infected. (Exp. 223) April 3, 1905, re-inoculated with 5 c.cm. blood from *Yolo* showing four trypanosomes to a cover; accidentally killed November 4, 1905. No trypanosomes nor signs of trypanosomiasis seen in it or in a rat sub-inoculated from it.

Rat (EXPERIMENT 135), July 6, 1904. Inoculated with buffalo blood; never infected. (Exp. 224) April 3, 1905, re-inoculated with 5 c.cm. blood from *Yolo* showing four trypanosomes to cover; never infected. Died of skin disease, October 13, 1905.

Rat (EXPERIMENT 139), July 27, 1904. Inoculated with 8 c.cm. blood from antelope; never infected. (Exp. 169) December 27, 1904, re-inoculated with 7·5 c.cm. of mixed blood and citrate solution from *Moiuma* showing four trypanosomes to cover; never infected. March 9, 1905, re-inoculated with 5 c.cm. of blood from *Keowkosiko*. March 21, 1905, re-inoculated with 5 c.cm. of blood from *Zilani*. April 14, died from mechanical effects of inoculation with large amount of blood; no autopsy; never infected.

Rat (EXPERIMENT 140), July 17, 1904. Inoculated with 8 c.cm. of blood from antelope; never infected. (Exp. 168) Dec. 27, 1904, re-inoculated with 7·5 c.cm. of blood and citrate solution from *Moiuma* showing four trypanosomes to a cover; never infected. March 9, 1905, re-inoculated with 5 c.cm. blood from *Keowkosiko*. March 21, 1905, re-inoculated with 5 c.cm. blood from *Zilani*. April 3, 1905, re-inoculated with 5 c.cm. blood from *Yolo*. April 13, 1905, re-inoculated with 5 c.cm. blood from *Yolo*. Never infected* up to September 19, 1905, when it escaped.

Rat (EXPERIMENT 170), December 27, 1904. Inoculated with 2·5 c.cm. blood showing four trypanosomes to cover from *Moiuma*; FIVE TRYPANOSOMES TO COVER WERE SEEN IN ITS BLOOD next day and not again. March 9, 1905, re-inoculated with 4·25 c.cm. blood from *Keowkosiko*. March 21, 1905, re-inoculated with 5 c.cm. blood from *Zilani*. April 13, 1905, re-inoculated with 33 c.cm. blood from *Yolo*. THREE TRYPANOSOMES TO A FIELD were seen in its blood on April 21, and not again. Died February 5, 1906; no trypanosomes seen at autopsy; all organs normal save spleen which was slightly enlarged and fibrous.

Puppy (EXPERIMENT 219), April 3, 1905. Inoculated with 10 c.cm. of blood from *Yolo*. April 5, 1905, re-inoculated with 10 c.cm. of blood from *Zilani*. April 13, 1905, re-inoculated with 125 c.cm. of blood from *Yolo*. Under observation until stolen at Boma. Trypanosomes never seen.

Monkey (*Cercopithecus schmidti*) (EXPERIMENT 220), very young. April 3, 1905, inoculated with 7·5 c.cm. blood from *Yolo*. April 5, 1905, re-inoculated with 5 c.cm. blood from *Zilani*. April 13, 1905, re-inoculated with 12·5 c.cm. blood from *Yolo*. Died of sunstroke May 10, 1905; parasites never seen. No trypanosomes or signs of trypanosomiasis at autopsy.

* See charts for number of parasites present in blood inoculated.

Through the kindness of Commandant Verdick the four infected cattle which remained when we left Kasongo were kept apart from the herd at Kasongo. After our departure the animals were left quiet, and there was a consequent amelioration in their condition. While we were there they were driven twice daily from their kraal to our laboratory and back—a distance of about one and a quarter miles. The fatigue of these journeys and the time last through them from grazing had certainly an adverse influence. *Moiuma* and *Keowkosiko* both calved. All the animals were in excellent condition in September, 1905. They were then sent to Lokandu, where there is a great deal of cattle trypanosomiasis and where *Glossina palpalis* is far from rare, in order to ascertain whether they would resist exposure to re-infection. (It must be remembered that the pasturage at Lokandu is not nearly so good as at Kasongo). The steer, three cows and two calves reached Lokandu on November, 23rd, 1905, in excellent condition. A calf died in December after twenty days of manifest diarrhœa. One of the adults died after a "slow wasting" and great weakness early in 1906. In August, 1906, two of the four remaining animals were very thin and seemed ill; the remaining two are still under observation. The Chef de Secteur at Lokandu, in conveying this information, reports that the "health of the herd at Lokandu is excellent."

The cattle at Kasongo graze on a wide plain intersected by several streams which have occasional clumps of bushes along their banks. *Glossina palpalis* are very, very rarely found about the grazing ground, and large game practically never comes near it.

TSHOFA, May 15th, 1905. There was formerly a herd here of some fifty cattle, which was intended to be used for supplying bullocks for work on the waggon road between Tshofa and Pania Mutombo. As the animals were not doing well, all, save fourteen which were obviously ill, were sent to Lubefu. Of that fourteen only eight remain; two were killed by a leopard, four died of a chronic disease characterised by general loss of condition and constant diarrhœa, with straining and small watery stools; there were no oedemas nor lack of appetite. Between June, 1901, and May, 1904, about thirteen animals were said to have died here of this disease. Almost all the calves born or brought here have died of it, and the

only animal present which was born here is affected by it (trypanosomes seen).

The majority of the cattle brought to Tshofa came from Lusambo. Cattle have been sent from the Tshofa herd in 1902 to Kisenga and in 1904 to the Tanganyika, Moero and Lubile districts. Three out of five animals whose gland juice and blood were examined had trypanosomiasis.

There was good pasture in open country at Tshofa, but *Glossina palpalis* and large game were very plentiful.

CABINDA, June 3rd, 1905. Before the advent of Europeans, some of the important chiefs in this neighbourhood had small herds of ten to twenty cattle. Cattle did not do well; many died of diarrhœa, and there were no large herds. At present a few head are kept here for vaccine making. They come from Portuguese West Africa by way of Angola and from as far South as Dilolo. We are told that the Dilolo natives buy cattle from the Zambesi valley. The blood and gland juice of six animals were examined once; trypanosomes were not seen.

LUSAMBO, June 21st, 1905. Nearly all the Lusambo cattle come by way of Malange and Luluabourg from Portuguese West Africa; formerly herds of a hundred head were brought in by this route to be exchanged for slaves. It is said that the animals were used for food and were not kept for breeding. The records of the Lusambo cattle are very badly kept,* but deaths from disease seem to have been rare among them. Six of the thinnest animals were chosen and their blood and gland juice were examined once. Trypanosomes were seen in one of them. About six cattle were sent from this herd to Kutu on Lake Leopoldville II in 1902-1903; all died within a short time. It is said that the cause of death was trypanosomiasis.

* This fault was a very usual one. The system of brands used for marking the cattle was also inadequate. It was consequently often impossible to identify an animal or to learn its past history. As a result it was often impossible at a station to estimate the incidence of disease in the past or to determine the origin of infected animals. It is evident that much valuable information would be obtained through the institution of careful systems of recording and identifying cattle.

III. OBSERVATIONS ON TRYPANOSOMES IN OTHER DOMESTIC ANIMALS

Horses, Mules and Donkeys: Comparatively few of these animals have been brought into the part of the Congo Free State visited by us. Some were seen in apparently good condition at the following places: various localities in the Lower Congo, Leopoldville, Eala (three horses and a foal from Dakar present for fifteen months, not worked), Coquilhatville (donkeys present four to five years), Bamamia, four donkeys (one present for eight years), constantly worked. Nouvelle Anvers, a few horses here were in good condition when we saw them in 1904 and were still well in June, 1905. Umangi (three horses, a mare from Teneriffe present for five years), Lisengi (two horses present for six years). At Nyangwe there is a small drove of donkeys. None of the above animals were examined. At Kasongo no trypanosomes were seen in two mules and a donkey; these three animals had journeyed considerably in the Eastern part of the Congo Free State.

In July, 1904, Commandant Sillye bought two stallions near Dakar in Senegal. They were then in excellent condition, and were six and twelve years old. He brought them to the Congo and took them on a hard journey from Kasongo almost to Lake Tanganyika; from this journey they returned unloaded to Kasongo, where we saw them on January 14, 1905, after they had had a fortnight's rest. Both animals were thin, but this was thought to be due to fatigue and bad pasturage. There was (Fig. 4), as in the Gambian horses (6), slight abdominal fulness without definite oedema. Both had rough coats from lack of grooming and were infected with ticks, fleas, chiggers, trombidium larvae and numerous bots. In addition both showed the weals of many recent fly-bites. If the animals were left to themselves they dropped their heads and certainly seemed to lack vigour.

The temperature of both was about 101° F. and their glands were slightly enlarged. The only sign of oedema ever noticed in either horse was a swelling of the scrotum in one in October, 1904. The conjunctivae were pale and watery in both. Trypanosomes were present in good numbers in the blood of both horses. The animals were relieved of the bots, chiggers, ticks and trombidia (lotion of bichloride of mercury) and were given arsenic in the form of Fowler's solution. At the end of January they reached Romee, where they were turned to pasture and the arsenic was discontinued. For two months both put on flesh. Then the older one became rapidly thinner and very weak; the appetite remained good. It died April 17, 1905, after a very profuse perspiration (6). On September 14, 1905, the remaining horse, though comparatively thin, seemed in fair health and fed well. The scrotum was often swollen.

Inoculations.—Rat (Exp. 180). Inoculated January 14, 1905, intraperitoneally with a mixture of blood, from the older of the two stallions, and citrate solution, showing 15 living trypanosomes to a cover. Parasites appeared in its blood next day and were never absent until it was accidentally killed on March 21.

Autopsy.—Spleen much enlarged (6·25 × 1·5 × ·75 cm.), soft, friable; glands all slightly enlarged.

Rat (Exp. 181). Inoculated as in Exp. 180. Parasites appeared January 31; constantly present till it died of the disease September 24, 1905.

Guinea-pig (Exp. 179). Inoculated as in 180; not infected. March 10, re-inoculated with 1·5 c.cm. blood from Exp. 180 showing 30 trypanosomes to field; infected in five days. Parasites constantly present till death from sunstroke, June 18, 1905.

Autopsy (immediately after death).—Dependent part of abdominal wall firm, water-logged; vulva oedematous; spleen soft (5 × 2·5 cm.); glands not enlarged. Animal well nourished.

Guinea-pig (Exp. 178). Inoculated as in Exp. 180; not infected. Re-inoculated as in Exp. 179. Trypanosomes in blood in six days, and constantly present until death from the disease on September 17, 1905.

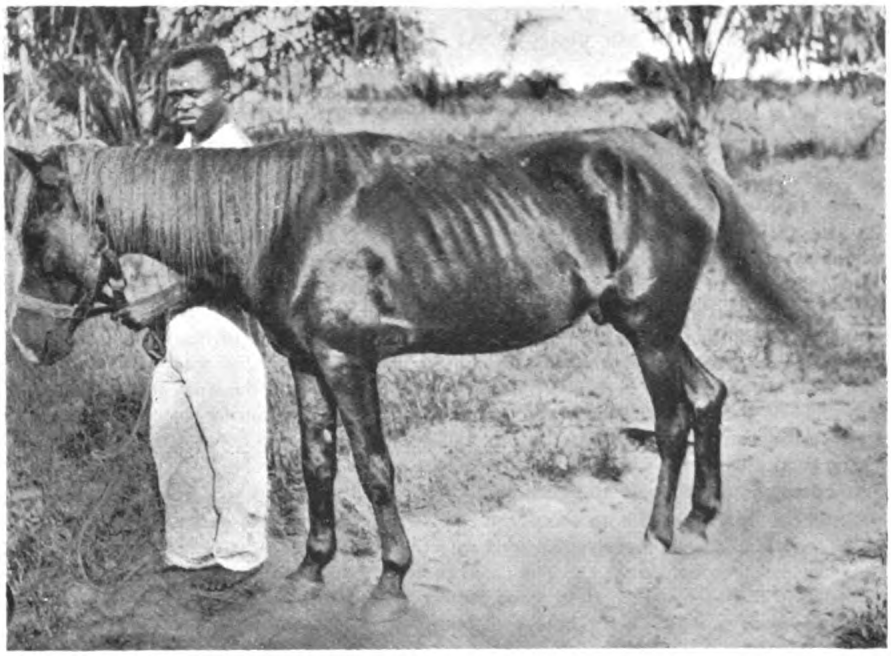


FIG. 4. STALLION infected at Kasongo. Note slight fulness of abdomen.

A donkey about seven years old, also belonging to Commandant Sillye, was examined on January 14th, 1905. This animal probably came originally from Uganda. Since 1902 it had been constantly worked and had just returned from the same journey as the horses. It was very fat and apparently in the very best of health. A few small "tadpole-like" trypanosomes were seen in its blood. Although hard

worked it remained in apparently robust health until the end of June, 1905, when it was accidentally killed.

Goats seem to thrive almost everywhere in the Congo.

Sheep are present in few places, but they usually do well.

None of the score of goats and sheep examined by us were infected with trypanosomes. Broden reports trypanosomiasis in sheep at Leopoldville.[3]

IV. TRYPANOSOMES IN BIG GAME

MSWATA.

Antelope (adult *Tragelaphus scriptus*), very scanty stumpy trypanosomes in blood; retro-peritoneal glands deeply haemorrhagic, otherwise apparently healthy.

Rat (Exp. 130), June 26, 1904. Inoculated with 3·5 c.cm. blood. Trypanosomes seen in its blood only on July 5. Died (cause, exposure ?) July 24; no trypanosomes or signs of trypanosomiasis at autopsy.

TSHUMBIRI.

Buffalo (*Bos nanus*, two adults). Both fat, and save for a caseous retro-peritoneal gland in one, apparently healthy. No trypanosomes found in blood.

Four rats (Exps. 132, 133, 134, 135) (see Inoculations at Kasongo, page 243) were inoculated with from 5 to 4 c.cm. of blood; none of them became infected.

COQUILHATVILLE.

Antelope (young *Cephalophus dorsalis* ?), no trypanosomes in blood.

Inoculated two rats (Exp. 139, 140) (see Inoculations at Kasongo); never became infected.

STANLEYVILLE.

Antelope (adult *Cephalophus dorsalis* ?). Gland juice and blood examined; no trypanosomes seen. Prescapular gland, enlarged ?, 4 by 3 by 2 cm., cortical congestion of it and retro-peritoneal glands.

LOKANDU.

Two antelopes (*Cephalophus dorsalis* ?) gland juice and blood examined; no trypanosomes seen. The blood and gland juice of an adult buffalo were examined and with its blood a rat (Exp. 133) was re-inoculated; trypanosomes were not found.

KASONGO.

Three antelopes (adult males, *Tragelaphus scriptus*).

1. Shot four hours previously, no trypanosomes in gland juice or blood. Prescapular gland 6 by 3 cm., iliac glands haemorrhagic.

2. Shot three hours previously. *Trypanosoma theileri* in blood. Inoculated rabbit and two guinea-pigs with blood; none became infected with any trypanosome.

3. Shot three hours previously; no trypanosomes in gland juice, many in blood. Abdominal glands much congested and enlarged.

Inoculated from Antelope No. 3.—Guinea-pig (Exp. 188). Inoculated January 28, 1905, with 1·5 c.cm. of blood showing three trypanosomes to field; not infected. April 19, re-inoculated with 2 c.cm. mixed citrate solution and blood from Exp. 189 showing one trypanosome to field; June 19 infected; trypanosomes were constantly present until animal's death from pneumonia on August 30, 1905. Spleen 3 × 1·5 × ·75 cm. normal; glands congested, not much enlarged.

Guinea-pig (Exp. 189). Inoculated with 2·5 c.cm. of blood as in Exp. 188; infected February 6, parasites constantly present in large numbers until death from pneumonia, August 30, 1905. Spleen large (6 × 3 × 1·5 cm.), soft, friable, congested; glands enlarged and congested. No trypanosomes were seen in blood just before death or in organ juices; present in gland juice.

Rat (Exp. 190). Inoculated with 2·5 c.cm. blood as in Exp. 188; not infected. April 19, re-inoculated with 2·5 c.cm. mixture as in Exp. 188; April 25, trypanosomes present, but were not again seen. Animal escaped June 19, 1905.

Rat (Exp. 191). Inoculated and re-inoculated (April 19) as in Exp. 190; infected May 1. Parasites constantly present until it died of pneumonia September 19, 1905.

Young guinea-pig (Exp. 230). Inoculated April 19, 1905, with 2 c.cm. blood from Exp. 189; infected May 6. Many parasites constantly present until death from the disease, July 6, 1905.

On the journey from Kasongo to Lusambo the blood of the following animals was examined by cover-slip preparations: four *Tragelaphus scriptus*, one *Canis anthus*, six *Hippotragus equinus*, one *Bos nanus* (ten antelopes, a jackal and a buffalo). Trypanosomes were seen in none of them.

V. MORPHOLOGY OF THE TRYPANOSOMES

(*a*) *Trypanosomes of cattle*

The trypanosomes seen in stained preparations of blood from the cattle, horses, antelopes and their sub-inoculations, are so nearly identical that one description will suffice. Slight variations and peculiarities will be mentioned separately.

Three types of trypanosomes were found corresponding closely to the description of *Trypanosoma dimorphon* as given by Dutton and

Todd.[6] The first of these is a very short form resembling the "tadpole" form. It measures between 8 and 14μ in length (most commonly 11 to 12·5) by from 0·9 to 3·4μ in breadth (average 1·5). In this type the trypanosome is widest slightly posterior to the nucleus, and then gradually tapers towards the anterior end. The posterior extremity presents all gradations in shape from acute to rounded; most commonly, however, it is rounded. The anterior extremity is effilated. The blepharoplast is small, round or oval, and is situated close to the posterior end and at one edge of the trypanosome. In most of the parasites the undulating membrane could not be seen; in those in which it was visible, it was extremely narrow. The thickened edge of the undulating membrane is poorly marked and in all cases ends abruptly with the anterior end of the trypanosome. A free flagellum is absent. The nucleus lies in the middle third, and as a rule occupies the whole width of the body. It is oval, and either stains quite homogeneously or is composed of a varying number of darkly-staining chromatin granules (5 to 17) arranged in no definite manner. These granules were not joined by connecting threads as far as could be made out. In only a very few cases could a structure suggesting a karyosome be seen. The cytoplasm stains a light blue and appears to be quite homogeneous. It was usually devoid of granules, but in some cases contained them.

The second type is allied to the "stumpy" form.[6] It measures from 15 to 20μ in length (average 16μ) and from 1·5 to 2·5μ in breadth (average 2μ). It also lacks a free flagellum or, at most, has an exceedingly short one. In other respects it does not differ materially from the description given above.

The third type resembles the "long" form.[6] It measures from 20 to 30μ in length (average 23 to 27μ) by from 1·2 to 2μ in breadth (average 1·5 to 1·7μ). The nucleus is rather elongated in the direction of the long axis of the body and as a rule stains quite intensely and homogeneously. This form has a fairly long free flagellum which measures from 2·5 to 6·5μ in length. The posterior extremity is rather acute and the blepharoplast is situated farther away from it than in the other two types. In other respects it is comparable to the two other varieties.

EALA.

The trypanosomes observed, with one exception, are of the "long" type, with a well-defined free flagellum. The exception measured 14μ in length by ·9μ in breadth, and conforms in all respects to the "tadpole" type. In these parasites the blepharoplast was of comparatively large size. Parasites of the "long" type in the process of longitudinal division were seen in preparations of the peritoneal fluid. In fresh preparations this trypanosome moved so rapidly that it was extremely difficult to keep it in the field of the microscope. By its rapid movement this parasite recalls the description of *Trypanosoma vivax* by Ziemann.[20] We are, however, satisfied that it is the "long" form of *Trypanosoma dimorphon*.

NOUVELLE ANVERS.

The parasites conform to the description of the "tadpole" variety. One trypanosome of the "stumpy" variety, measuring 19·2 by 1·9μ, was also found. In the one sub-inoculation the trypanosomes did not show any changes.

ROMEE.

The parasites are of the extremely short "tadpole" variety. In the direct sub-inoculations the parasite preserved the same characteristics with considerable constancy. In two cases (Exps. 148 and 151), in addition to the very short forms, trypanosomes corresponding to the "stumpy" forms made their appearance. The same holds good with regard to the second passage. This fact is of some importance. Dividing forms of the usual longitudinal type were observed in all the sub-inoculations.

LOKANDU.

Trypanosomes of the first two types (the "tadpole" and "stumpy" forms) were found in these cattle. The short "tadpole" forms were very much more common than the "stumpy" type.

SENDWE.

The parasites agree in all respects with the "tadpole" type. The examination was rather restricted, however, as the parasites were very scanty and only a few preparations were available.

KASONGO.

In the cow, *Moiuma* (No. 33), the trypanosomes are of the third, the "long," type, and possess a well-marked flagellum. In many of

these parasites a large vacuole was seen immediately in front of the blepharoplast. In this parasite the blepharoplast was comparatively large, and recalls the trypanosome seen at Eala.

In the other cattle, *Zilani* (No. 8), *Yolo* (No. 9), and *Keowkosiko* (No. 60), the trypanosomes are of the "tadpole" variety. In Yolo, however, one parasite was seen of the "long" type with free flagellum and comparatively large blepharoplast. The further history in sub-inoculations of the trypanosome derived from *Yolo* is of great importance in its bearing on the identity of these cattle trypanosomes. In the rat (Exp. 222), inoculated from Yolo, all three types were found. Of these the "stumpy" type, measuring from 15 to 18μ in length, was most abundant. In the subsequent passage this differentiation became more pronounced. The very short forms, measuring up to 14μ in length disappeared, but the other two types persisted. The number of "long" forms increased with the progress of the disease, as was noted by Dutton and Todd in the case of *Trypanosoma dimorphon* ([6], page 37).

The remaining two types can still easily be distinguished in experimental animals. The "long" form is more plentiful, is slender, and has a rather acute posterior extremity. The flagellum is fairly long and is actively motile, whipping from side to side with great vigour. The movements of the whole trypanosome are active and progressive. The trypanosomes of the "stumpy" type are rather broad and have a blunt posterior extremity. They move slowly, twisting round on themselves, then straightening out again, and so on. As a rule they maintain the one position and do not move from place to place. The undulating membrane moves sluggishly, and there appears to be no, or, at most, a very short, free flagellum. The body of the trypanosome is itself contractile. When buried between corpuscles the movements of both forms become appreciably slower.

(*b*) *Trypanosomes of the Kasongo stallions*

In the younger horse the parasites were identical with the "tadpole" forms as observed in the cattle. Unfortunately, no sub-inoculations were made from this animal.

In the second horse, "Toul," the trypanosomes were of the second ("stumpy") and third ("long") types as seen in the cattle. The "long"

forms with free flagellum were in the majority. In both types the cytoplasm often contained small reddish granules, which were most frequently observed in the posterior half of the body. In some thirty sub-inoculations with this strain the trypanosomes of both types have been constantly present. At the inception of the disease the " stumpy " forms are much more abundant, but as the affection proceeds the " long " type assumes the preponderance. ***In the monkey sub-inoculated a few of the " tadpole " forms were observed.***

In fresh preparations the two types can be very easily distinguished. The long, slender form has a long flagellum and moves very rapidly, lashing in and out among the corpuscles. It progresses for a short distance, then stops, and in a few moments moves on again. It often moves fairly rapidly across the whole field. Occasionally it adheres to a corpuscle, and after twisting and lashing about for a short time breaks free and moves off. While the parasite progresses with the flagellar end in front as a general rule, it occasionally moves backwards for a short distance. The second form is broader and shorter than the first, and is much more sluggish in its movements. These consist chiefly of doubling and twisting of the trypanosome on itself. It does not progress far, but remains pretty constantly in the one spot. The undulating membrane and flagellum are very poorly developed in this variety. In contradistinction to Dutton and Todd,[6] dividing forms, of the usual longitudinal type, were seen in this " stumpy " type.

(*c*) *Antelope trypanosomes*

In the antelope (*Tragelaphus scriptus*) killed at Mswata trypanosomes corresponding to the " tadpole " type were seen, but were very scanty. In the one sub-inoculation the parasites retained this type. Dividing forms of the usual longitudinal type were observed.

In the antelope at Kasongo the trypanosomes were of the second (" stumpy ") and third (" long ') varieties as found in the cattle. In the sub-inoculated animals these two types persisted; the stumpy forms being in the majority in the preparations examined. It was impossible to study this parasite fully, since the animal in which it was brought to England was accidentally killed and the strain was lost.

VI. ANIMAL REACTIONS OF THE TRYPANOSOMES

(*a*) *Trypanosomes of cattle*

The slight infectivity for laboratory animals of the trypanosomes in the majority of the cattle found infected is very striking. It is only at Romee that experimental inoculations were at all successful. No morphological variation in the parasites was noted to accompany this difference in virulence. All the parasites seen agree with the types already described. The results of inoculation of various laboratory animals with the trypanosome obtained from *Yolo*, Steer 9 at Kasongo through rat (Exp. 133) are briefly given.

MONKEYS (2).

Two monkeys, a *Macacus rhesus* and a *Cercopithecus* sp ? were inoculated intraperitoneally with small quantities of infected blood (0·5 and 2 c.cm. respectively). Trypanosomes were never seen in the *Macacus*, although it was carefully examined every day. It died 72 days after inoculation, from general tuberculosis. Parasites were found in the *Cercopithecus* on the second day after, but were only present in the peripheral circulation for two days ; they then disappeared and were never seen up to the time of the monkey's death, some six weeks after inoculation. The monkey had a rise in temperature to 104° F. while trypanosomes were present in the blood, but afterwards did not have a recurrence of the fever. A rat was sub-inoculated from this monkey on the day of its death with 11 c.cm. of almost pure blood, but has not become infected up to the present, 80 days after inoculation.

RABBITS (3).

The incubation period varies considerably, from four to sixty-nine days, the usual time being about three weeks. The disease tends to be very chronic in these animals, as is strikingly exemplified by the following experiment :—

A rabbit was inoculated subcutaneously on May 3, 1906, with 4 c.cm. of infected blood and became infected 25 days later. The trypanosomes were present in scanty numbers for a week and then disappeared from the peripheral blood. Since June 4, 1906, they have never been seen, although the rabbit has been under continuous observation. On July 31, two rats were sub-inoculated from the rabbit, and both became infected after an incubation period of between two and three weeks. On November 12, 1906, another rat was sub-inoculated from the rabbit and became infected two weeks later. Finally another rat was sub-inoculated on January 30, 1907, and became infected after a prolonged incubation period of 47 days. It will thus be seen that the blood is still infective after the lapse of eleven months. It is said that unfavourable conditions, as lack of food, render animals particularly susceptible to trypanosomiasis and may cause a declaration of latent infection. In order to test this hypothesis this rabbit was placed on a very restricted diet from December 1, 1906, to the middle of February of this year. During this time it decreased in weight from 2,790 grm. to 2,075 grm. Trypanosomes were never seen in the blood, however, and as stated above a rat sub-inoculated at the end of January became infected only after a prolonged incubation. The temperature remained normal throughout.

Rabbits do not show any symptoms, but remain quite healthy and well-nourished. The trypanosomes are never very plentiful, and usually show periodicity. Two rabbits which died had extensive coccidiosis, and their death in less than four months after inoculation was probably due to this secondary infection.

Post-mortem.—The spleen is slightly enlarged and rather congested. The lymphatic glands are usually quite pale, firm, and only very slightly enlarged.

GUINEA-PIGS (3).

The incubation period is prolonged and the disease is also of long duration—up to 139 days. The animals remain quite healthy and the temperature does not rise during the disease. The trypanosomes are rather scanty at first, but before death occurs they increase in number and may finally be fairly plentiful. At the autopsy of those dead of the disease the spleen is found to be enlarged and rather soft. The lymphatic glands are increased in size to a slight extent and are quite pale in colour.

RATS (14).

Rats are rather susceptible to the disease. The incubation period is short, from five to fourteen days. It is interesting to note that in the rat sub-inoculated directly from the steer (*Volo* No. 9) trypanosomes were not seen until the lapse of *249 days*. When once established, the disease runs a chronic course as a rule, lasting for from 40 to 254 days. The usual length is about 80 days. The parasites are scanty, but usually increase in numbers before the animal's death. Periodicity is marked; the parasites may be absent from the blood for two or three weeks. The mode of inoculation has no influence on the incubation period. Large doses may cause the parasites to appear a little sooner than they otherwise would, but rats inoculated with such doses do not die any more quickly than those inoculated with small doses of infected blood. The quick passage of the trypanosome through a succession of rats (six) did not increase the virulence in any way.

Post-mortem.—The spleen is enlarged, congested and rather firm. The lymphatic glands are slightly larger than normal, and are quite pale and firm. The other organs show no changes.

MICE (2).

As in rats, the incubation period is short, about four days, and the disease is chronic. One mouse is alive 219 days after inoculation and still infected. The trypanosomes are always scanty, and may be absent from the peripheral blood stream for many days—78 on one occasion. The post-mortem changes are similar to those found in rats.

It will thus be evident that most of the common laboratory animals are susceptible to the infection. Mice and rats are most easily infected, guinea-pigs least. The disease is chronic and lasts for a comparatively long time. It produces few or no symptoms and the trypanosomes are usually present in but small numbers. The strain is not very virulent, and resembles in this the strain derived from the horse.

(*b*) *Horse trypanosomes*

The animal reactions of the strain derived from the second horse, *Toul*, are epitomised briefly below.

MONKEYS (2).

Two monkeys were inoculated, a baboon (*Papio anubis*) and a *Macacus rhesus*. The baboon, inoculated intraperitoneally with 5 c.cm. of blood showing one trypanosome in from one to five fields, never became infected. The *Macacus*, inoculated subcutaneously with 2 c.cm. of citrated infected blood, became infected after an incubation period of six days. At the same time the temperature rose to 105·4°. Unfortunately the further temperature chart is not of much value, as the monkey developed tuberculosis. The trypanosomes showed periodicity in their appearance and disappearance in a marked degree. The parasites were scanty at first, but became more numerous in the course of a couple of days and were afterwards present in fair numbers. The monkey died 107 days after inoculation, and at the autopsy the spleen and liver were found to be filled with tubercles. On the other hand, the lungs appeared quite normal. The lymphatic glands were not enlarged.

DOGS.

One puppy was inoculated subcutaneously with 1 c.cm. of citrated blood from a rat, and twelve days later trypanosomes were found in the peripheral circulation. The parasites have been periodic in their appearance and have never been present in large numbers. At the time of the first appearance of the parasites there was a slight rise in the dog's temperature, but since that there have been no further increases corresponding to the appearance of trypanosomes in the peripheral circulation. The dog is quite healthy and presents no symptoms whatever. It is still living, 76 days after inoculation.

RABBITS (5).

Five rabbits have been inoculated with varying doses of infected blood. The incubation period varies from eleven to twenty-nine days; most usually it is about two weeks. The trypanosomes are periodic in their presence in the peripheral blood and are usually rather scanty just before an increase in numbers. One rabbit only, showed in a marked degree the symptoms usually found in trypanosomiasis—emaciation, loss of hair with following ulceration of the skin, oedema of the external genitals and the base of the ears, increased secretion from the nostrils, conjunctivitis, haemorrhages into the anterior chambers of the eyes, panophthalmitis and blindness. The animal also became very anaemic and there was a diminution in both the number of red cells and the percentage of haemoglobin. All the others showed no symptoms. The disease is very chronic in rabbits, and lasted 173 and 178 days in the two which died from the disease. Two other rabbits died early in the disease from intercurrent affections. The fifth rabbit is still alive, 76 days after inoculation. There was no marked rise of temperature when the rabbits first became infected.

At the post-mortems of the animals dying of the disease enlargement of the spleen and lymphatic glands was the most noticeable feature. The lymphatics were pale in colour and were rather firm in consistence. The spleen was congested and rather soft.

GUINEA-PIGS (10).

Guinea-pigs appear to be most resistant. The incubation period varied between seven and fifty-two days, six weeks being the average time. The disease, when once established, is of long duration; death ensued in from 46 to 352 days, about 90 days being the average. Two are still alive, 76 days after inoculation. Trypanosomes are still present in both. In a few of the guinea-pigs the parasites were continuously present from the date of their first appearance in the peripheral circulation until the animal's death. They fluctuated in numbers, however. In the majority periodicity was noticeable. Occasionally parasites were absent from the peripheral blood for long periods, e.g. 34 and 67 days.

No symptoms of any sort are noticeable in guinea-pigs. The virulence of the strain was not increased by successive passages through three guinea-pigs; the incubation period and the duration of the disease were not affected. The temperature chart does not reveal any rise during the course of the affection. The trypanosomes are usually rather scanty, but before the death of the animal they may increase in numbers until they are fairly plentiful.

At the autopsy a more or less marked enlargement of the spleen was seen. The organ was congested and rather soft. The lymphatic glands, as a rule, were unchanged, though in a few instances they were slightly enlarged and in one case haemorrhagic. The other organs appeared normal.

RATS (10).

Rats are rather susceptible to the disease. The incubation period varies between one and forty-one days; as a general rule it is about four to six days. The duration of the disease varied from nine to 253 days, usually about 70. In the acute cases the parasites are present continuously until death, and are usually fairly numerous. In the chronic cases periodicity is observed and the trypanosomes, when present in the peripheral circulation, are usually in small numbers. Increase in the virulence of the strain was not obtained by passage through a succession of four rats. Sub-inoculations made from other experimental animals became infected in approximately the same time.

Post-mortem.—The spleen is enlarged and is congested and soft in consistence. Haemorrhagic infarcts were seen in one case. The lymphatic glands may or may not be enlarged. The enlargement is usually slight when it is present. The glands are pale and rather firm. The other organs normal. In one case subpleural petechiae were observed.

MICE (2).

As in rats, the incubation period is short, in our experiments about two days. The disease is chronic and lasts for months—as long as five. When first present the trypanosomes are very scanty. They disappear in a few days and may not be seen again in the peripheral blood for weeks. In one case they were absent for fourteen. They reappear before death and increase in number until the animal succumbs.

Post-mortem.—The spleen is very much enlarged, congested, dark purple in colour, and rather soft in consistence. The lymphatic glands are not enlarged. The other organs appear normal.

In general, the trypanosome is not very virulent. The incubation period is prolonged and, while the disease has *always* terminated fatally, death only ensues after the lapse of a comparatively long interval. The low degree of virulence is also manifested by the lack of symptoms and the absence of heightened temperature in the experimental animals. Efforts to increase the virulence by successive passages through any one species of animal (four passages) or through an alternation of animals, e.g., guinea-pigs, and rats (three of each) were without result. The parasites are scanty, as a rule, and in the more chronic cases exhibit periodicity in a very marked degree.

In the following sections the cattle trypanosomes in the Congo Free State are treated as a whole.

VII. DIAGNOSIS

An absolute diagnosis, of course, depends upon the demonstration of the parasite. The charts of the Kasongo cattle show that every means may frequently fail to show the parasites in infected animals. Of all the methods gland puncture was the most successful. In thirty consecutive examinations, at which parasites were detected either in the gland juice or in the blood, gland puncture was successful twenty-nine times, the examination of cover-slip preparations of blood thirteen times; on one occasion trypanosomes were present in the blood and were not seen in the gland juice. The same precautions must be taken in puncturing the glands of cattle as in those of men.[5] The records of the animal inoculations (particularly at Kasongo, page 243) show that the animals we employed frequently remained uninfected after the inoculation of large quantities of blood containing living parasites. As a diagnostic test the sub-inoculation of laboratory animals was therefore often less sensitive than the examination of cover-slip preparations of blood.

If we except the cattle at Romee (seven laboratory animals, all infected), the bovine trypanosomes (i.e., first passages) were remarkable for their slight virulence in direct inoculation. Of twelve rats, four guinea pigs, two rabbits, two monkeys and four dogs inoculated at various stations, only one rat ever became infected. It is noteworthy that at present sub-inoculations into ordinary laboratory animals of the strain derived from this rat are always successful. The virulence of this trypanosome in direct inoculations therefore seems to have increased for the animals employed. The original strains of trypanosomes obtained from the antelope and from Commandant Sillye's stallion at Kasongo were much more virulent in the first passages.

No idea was formed of the absolute efficiency of gland palpation as a diagnostic test. In the animals found to be infected the superficial lymphatic glands were usually considerably enlarged; some of them, not advanced cases, however, had no evident glandular enlargement. Many of the animals examined once, and not found to be infected, had glands as large as, or larger than, those seen in the infected animals (this was especially so at Kasongo).

In the Gambia[6] it was noted that a heightened temperature furnished a valuable indication of possible trypanosome infection in horses. Charts I, II, III, IV and V fully confirm this observation; but a high temperature observed once in an animal just thrown after a struggle in the sun has but little value. In such circumstances temperatures of from 101·5°F. to 103°F., or more, were noted in

animals carefully examined for parasites without result and believed to be healthy.

The enormous difficulty of demonstrating the presence of parasites in animals known to be infected is well illustrated by the observations at Kasongo. The importance of a well-kept temperature chart is shown by *Liboko* (Chart IV), whose temperature was constantly elevated, although parasites were seen but once.

Animals suspected of trypanosomiasis for any reason should be isolated; their temperature should be regularly and quietly taken; their gland juice and blood should be frequently examined, and laboratory animals should be sub-inoculated from them with doses of 10 c.cm. of blood. An examination of this type extending over some months is necessary before an animal can even tentatively be said to be uninfected.

VIII. SYMPTOMATOLOGY

The symptoms are given in the order of their importance. Fever is practically always present. Listlessness, with progressive weakness, emaciation and anaemia are usually present. A tendency to diarrhœa, with small frequent watery stools, and enlargement of superficial lymphatic glands* are frequently striking features. Oedemas and running from the eyes and nose were seen in only one or two instances. Haemorrhages, eye lesions, staring of the coat and loss of hair were not observed. The appetite remains good to the end. In the autopsies done the enlargement of the lymphatic glands was by far the most striking feature; the glands were either pale and very watery (particularly superficial glands) or congested and haemorrhagic (particularly abdominal or mediastinal glands). Oedemas were very rare, exudates were scanty, the spleen was not enlarged. We saw no petechial haemorrhages of serous membranes. On the whole, these findings are identical with those of Broden.[3]

IX. COURSE OF THE DISEASE

The disease is sometimes exceedingly chronic, and the infected animal may live for many months. (See Exps. 143, 146, 133, pages 238, 239, 244, and below).

If marked wasting continues death usually follows in both cattle and horses within a few weeks or three or four months. Occasionally an animal at this stage if well cared for will recover flesh, and the

*** We are unable to say exactly how long after infection enlarged glands become apparent. We are inclined to think that it may be a comparatively late manifestation.**

infection resumes the latent form. As is evident from the observations in many places, domestic animals may live an apparently normal life in districts where trypanosomiasis is rife; some infected animals may live for years in apparent health in such districts (see below).

The following observations are interesting in this connection. The history of Horse VI suggests that animals naturally infected with *Trypanosoma dimorphon* may sometimes recover, but are not immune to re-infection.

A stallion, Horse VI (Ref. 6, page 30), found naturally infected in the Gambia was brought to England. During two and a half years there were occasional rises in temperature, trypanosomes were occasionally seen, and its blood was infective to rats and mice (7). The temperature then became more regular, its blood was no longer infective, and parasites could not be found. In October, 1905, the superficial lymphatic glands were just palpable and too small to be punctured. During the winter of 1905, the animal became thin, &c., but trypanosomes were not seen. During the whole of 1906 the animal was healthy, its temperature was normal, and trypanosomes were absent from its blood. The glands remained too small to be punctured. From time to time inoculations were made into susceptible animals as follows:—

July 23, 1906. A white rat, inoculated subcutaneously with 7 c.cm. blood. As it had not become infected it was re-inoculated on August 14 with 18 c.cm. almost pure blood. It died on the 5th of September without having become infected. Cause of death was pneumonia.

October 23, 1906. A guinea-pig received 17 c.cm. of blood subcutaneously and died February 2, 1907. Trypanosomes were never seen, and at the autopsy all the organs appeared quite normal. On the same day a rat was inoculated with 10 c.cm. of blood. It died on November 5, 1906, from pneumonia; never infected.

January 15, 1907. A guinea-pig was inoculated subcutaneously with 20 c.cm. almost pure blood, and two rats with 20 and 15 c.cm. respectively. They have been carefully followed to date but have never become infected.

From all these points we believe this pony had recovered from a natural infection by *T. dimorphon*.

On the 21st of January, 1907, the pony was inoculated intraperitoneally with 5 c.cm. of infected citrated blood from a mouse infected with *T. dimorphon*. This was followed between the 6th and 9th of February by a rise in the animal's temperature, highest 102·5°, and again after a few days intermission to 103·2° on the 16th. With these exceptions the temperature has been since about 100°.

On February 16, two rats were sub-inoculated from the pony with 17 c.cm. of blood each. One of these became infected on the 6th of March and died six days later with numerous trypanosomes. The second rat became infected on the 15th of March and died on the 28th, also heavily infected. In both the spleen was very much enlarged. The trypanosomes were of the usual *dimorphon* type ("stumpy" and "long" forms).

In addition to the rise in temperature the pony lost its appetite and appeared sick. It lost weight for several weeks, but is now recovering. The blood was carefully examined every day but without result; trypanosomes have never been seen. The glands were palpated from time to time, but have shown no enlargement.

The following experiments indicate once more that animals inoculated in the laboratory with *Trypanosoma gambiense* may recover from the consequent infection. The connection between these and the preceding observations is evident. Their bearing on the question whether human trypanosomiasis is invariably fatal is most important.

I. A brown stallion was inoculated in the Senegambia (6) from a case of human trypanosomiasis. For the earlier history consult Memoir XVI (7) of this School. In October, 1905, the animal appeared to be quite healthy; no enlargement of the glands was present, and no trypanosomes could be found in the blood. Its temperature was always normal. Sub-inoculations from it were made as follows:

November 12, 1906. A guinea-pig was inoculated subcutaneously with 17 c.cm. of almost pure blood. It was accidentally killed on the 31st of December. Trypanosomes were never seen. On the same date a rat was sub-inoculated with 10 c.cm. Trypanosomes were never seen at any time. The rat died on February 4, 1907, from acute enteritis.

January 16, 1907. A guinea-pig and two rats were inoculated with 20, 17 and 20 c.cm. of pure blood respectively. The guinea-pig and the first rat are still alive and have never become infected. The second rat died on the 11th of February from an extensive skin disease.

This series of experiments pointed to the fact that the pony had entirely recovered from the infection by *Trypanosoma gambiense*. On January 21st it was re-inoculated subcutaneously with 7 c.cm. of citrated, infected blood, showing one trypanosome to one to five fields, from a guinea-pig infected with *Trypanosoma gambiense*. This, however, had no effect. The temperature has been taken twice daily for the past year; there has been no rise, even after the inoculation. The animal remained in good health; retained its appetite and did not lose weight. The pony was most carefully examined day by day, but trypanosomes were never seen.

On March 15 two rats were sub-inoculated, receiving 30 and 25 c.cm. respectively of almost pure blood. Up to date they have not become infected. On March 20 the pony was again inoculated intraperitoneally with 25 c.cm. of infected *gambiense* blood showing numerous trypanosomes. For a few days afterwards there was slight auto-agglutination of the cells, but trypanosomes have never been seen. As in the first instance, no rise of temperature or other morbid symptoms have been noticed. The experiments are still in progress.

II. A donkey was inoculated in 1904 (7). Since the fall of 1905, the animal has been in perfect health. No enlargement of the glands, or trypanosomes, have ever been noticed. On January 16, 1907, a guinea-pig (20 c.cm. pure blood) and a rat (20 c.cm.) were sub-inoculated, but up to date have not become infected. On January 21 the donkey was inoculated subcutaneously with 7 c.cm. of blood from a guinea-pig infected with *T. gambiense*. No rise in temperature or other symptom followed. For a few days the red cells showed auto-agglutination, but trypanosomes were never seen. On March 15 two rats were sub-inoculated from the donkey with 35 c.cm. of pure blood each. Both are still alive and have not become infected. On March 20 the donkey was re-inoculated intraperitoneally with 25 c.cm. of blood containing *T. gambiense* (one per field), but without result. No symptoms of any sort are apparent. The donkey is still under observation.

III. The cow inoculated by Thomas and Breinl (7) was under observation until June, 1906, when the animal passed out of our hands. It was in good health all the time. No trypanosomes were seen and the glands were not enlarged.

IV. A monkey (*Cercopithecus*) was inoculated May 17, 1904, with blood from a sleeping sickness case. She showed trypanosomes in her blood June 13; they were seen at intervals up to June 13, 1905, and not again. The animal died August 5, 1905, from miliary tuberculosis; no trypanosomes were seen at the autopsy. (This experiment is reported merely for its interest, as showing a chronic infection by *T. gambiense* in a monkey.)

X. CONCLUSIONS DISCUSSED

A. Cattle trypanosomiasis probably exists everywhere in the Congo Free State. Broden[3] is also of this opinion. Since large game and *Glossina* are practically omnipresent[1] (perhaps with the exception of high land about Lake Kivu) it seems possible that it should be so; at all events, trypanosomes were found in every herd examined save those at Nyangwe, Cabinda and Lusombo. That the cursory examinations made at these places might easily have failed to detect infected animals is certain (see charts).

B. The causative trypanosome is probably identical with *Trypanosoma dimorphon*. Three ways of identifying trypanosomes are at present recognised; all of them are liable to error. They are (1) The morphology of the parasite in infected animals and in cultures.* Cultures have been made of very few of the African trypanosomes, and were not used in the present instance. (2) Animal reactions. (3) Cross inoculations.

(1) Morphology. It is well known that the form and dimensions of all trypanosomes may vary more or less, but when an unknown trypanosome varies in exactly the same way as does a known trypanosome (compare the Congo parasites we describe and *Trypanosoma dimorphon*) their variations become a strong proof of identity. And this especially in the present case, since no trypanosome of domestic animals shows such wide divergencies in form as does *T. dimorphon*.

All the forms seen may be approximated to the three types of *Trypanosoma dimorphon* as described by Dutton and Todd.[6] At Eala, where the cow was suffering from an acute exacerbation of the disease, the "long" type were almost entirely seen, but one "tadpole" form was observed. At Romee, where the disease was chronic, only

* Koch's observations on *Trypanosoma brucei* and *T. gambiense* in the alimentary canal of *Glossina* suggest a realization of Schaudinn's proposal that an accurate classification and differentiation of various haematozoa would come with a fuller knowledge of their life cycles in either or both of their hosts. (Koch. Ueber die Unterscheidung der Trypanosomenarten. Sitzungsberichte der Königlich preussischen Akedemie der Wissenschaften, XLVI Sitzung vom Nov. 23, 1905.)

the " tadpole " type was found. In the sub-inoculations, however, " stumpy " forms made their appearance. At Lokandu the " tadpole " and " stumpy " forms were seen. At Kasongo " long " forms were seen on one cow, and " tadpole " forms in the others. In one of these a " long " form was also observed. The " stumpy " and " long " types was also seen in the antelope at Kasongo. Dutton and Todd described the " tadpole " as being present at the beginning of the disease, while the other forms of the parasite appeared later.[6]

It seems quite probable that the " tadpole " forms are really often present in chronically infected animals and that they give place to the " stumpy " and " long " forms, as the disease becomes more acute, in either the original host or in sub-inoculated animals. That this is a probable explanation is supported by several observations in the present paper. For example, the history of the parasite as observed in the case of *Yolo* (p. 253), where we have the change from the minute form to the other two types in the sub-inoculations. The similar change seen by Broden[3] in the case of his sheep and its sub-inoculation is of interest in this connection. Broden states that in the animals sub-inoculated from the cattle at Yumbi, the trypanosomes preserved the characteristics of those seen in the original animals. This observation seems to conflict with the explanation we have just given. He does not state definitely that forms with a free flagellum were absent, and from the fact that he compares some of his forms with the hyaline ones of Bradford and Plimmer which had a free flagellum, it seems possible that one was also present in some of the trypanosomes he saw. In this connection it is interesting to note that he observed only the " petite " forms in the cattle at Eala, whereas we saw both the " tadpole " and the " long " forms. The occurrence at the same time of trypanosomes of more than one of the types in several of our animals certainly also seems to point to the infection being caused by only one trypanosome rather than by two or more.

(2) Animal Reactions. The virulence of various strains of trypanosomes (in the laboratory) may vary greatly. The effects of a parasite on experimental animals can therefore only be taken as a general indication and cannot be accorded specific value. A parasite can only be said roughly to be highly, fairly, or slightly virulent. With *Trypanosoma dimorphon* the Congo parasites come under the second class.

The variation in virulence observed in the various strains of cattle, horse, and wild game trypanosomes found in the Congo is certainly not sufficient to suggest that more than one species of parasite was present.

(3) Cross Inoculation. This method depends upon the belief that an animal which has acquired immunity* to one trypanosome can only be re-infected by a trypanosome of a different species. We believe that this method is open to many fallacies, especially because of the enormous difficulties of applying it. (*a*, *b*, *c*).

(*a*) Experiments and observations of many authors show how extremely difficult it is to be certain that an animal has definitely recovered from a trypanosome infection. Cases of known infection often arise in which every means of demonstrating the parasite may fail.†

(*b*) For the same reasons an animal may become infected without the infection being detected.

(*c*) Our observations on Horse VI show that an animal may be twice infected by the same parasite.

Laveran (17) infected a rat with *T. gambiense* which had recovered a year previously from a previous inoculation. Martin (15) records somewhat similar experiences with a sheep and a goat. He states that "une même trypanosome peut revêtir des virulences variables et une forme attenuée ne vaccine point contre la forme exaltée," basing this on the work of Koch and Martini. This complicates the question of the identity of any one trypanosome, and shows that additional caution must be observed in placing too much reliance on the results of cross inoculations. It is altogether probable that the immunity in trypanosome infections is quite comparable to that seen in other protozoal diseases such as malaria and spirochaete infections, i.e., only a relatively active and not an absolute immunity is obtained.

Although most authors, in describing a newly-discovered trypanosome, have found it difficult to say to what species their parasite belongs, the majority of the trypanosomes, other than *Trypanosoma equiperdum*, reported in domestic animals in Africa, seem to be of the type of either *T. brucei* or *T. evansi*. For example,[20] Baleri as described by Cazalbou, Jinja cattle disease found by the members of the Sleeping Sickness Commission in Uganda, Kidei as described by Sanders, Aino found in Somaliland, El Debab found by Sergent in camels in Algeria, Mbori affecting animals in French Soudan, Souma described by Cazalbou, and the trypanosome described by Roger and Greffulhe in horses in Algeria.

* In our hands the serum reactions dependent upon such "immunity" have proved unreliable.

† This point cannot be too strongly insisted upon. It must be especially remembered by those engaged in experimental work on the treatment of trypanosomiasis.

The animal trypanosomes we found in the Congo approach *Trypanosoma dimorphon* most nearly in morphology and animal reactions. On these grounds we identify them with that species.*

Broden (3) believes that there are two varieties of trypanosomes, at least, in the Congo. One of these, corresponding to our "tadpole" forms, he calls *T. congolense*, while the other he assimilates with *T. vivax*, described by Ziemann in the Cameroun. At Leopoldville he found a cow with the two varieties of trypanosomes. From the fact that in the sub-inoculations he found only parasites corresponding to his small forms, he concludes that the animal was suffering from a double infection. It is possible that the explanation suggested above may apply in this case. Broden also remarks, in considering the identity of the small form, that Dutton and Todd were unable to discover their trypanosome in Gambian cattle. That their examinations were insufficient has been demonstrated by Captain A. G. Todd, A.V.C. (13), who has since found trypanosomes in these animals.

In the Soudan, Balfour (14) found a trypanosome in cattle which bears considerable resemblance to the "tadpole" type found in the Congo. Five animals—two monkeys, two rabbits and a dog—were sub-inoculated with small quantities of blood, but trypanosomes were never seen in any of them. As the author states, the number of experiments is too small for any dogmatic opinion to be given as to the non-susceptibility of these animals. It should be again noted here that of many animals inoculated from cattle by us only one rat became infected. Balfour's trypanosome was named *T. nanum* by Laveran. Is it not possible that further work on this parasite may show that it is merely the tadpole form of *T. dimorphon*? In addition to cattle, Balfour found mules infected, but in these the trypanosomes were frankly of the *T. dimorphon* type (stumpy and long forms). In these the long slender trypanosomes have a free flagellum.

In French Guinea Martin (15) found that most of the domestic animals suffered from one of two trypanosomiases. Of these, the most frequent was due to *Trypanosoma dimorphon*, and it is interesting to note that in a sheep the trypanosomes had a long free flagellum, thus once again bearing out Dutton and Todd's observations. In sub-inoculations, however, the flagellum was lost as in Dutton and Todd's strain of *T. dimorphon*. Another interesting point is that in cattle Martin noted that occasionally *only one* type of the parasite was present, most often the long form.

Quite recently Laveran (16) has described a trypanosome found in sheep coming from Ségou on the Upper Niger. This trypanosome presents many of the characteristics of *T. dimorphon*, but from the fact that a sheep which had recovered from the disease, and had afterwards resisted a second inoculation of the same strain, became *infective* when inoculated with *Trypanosoma dimorphon*, Laveran concludes that it is a separate species, and accordingly names it *Trypanosoma pecaudi*.

Trypanosoma congolense, *Trypanosoma nanum* and *Trypanosoma vivax*, at least, of other African trypanosomes, present marked affinities with *Trypanosoma dimorphon*. If they are identical the distribution of *Trypanosoma dimorphon* is very wide.

* At this juncture it is well to consider the variations in form and virulence described by Koch (20, page 121) to occur in *T. brucei*.

C. The transport of cattle from place to place should be forbidden as far as possible.

Apparently healthy animals may be infected with trypanosomes. It is easy, therefore, to understand the dangers which lie in moving cattle. An infected animal might easily introduce a new, and perhaps, therefore, a more dangerous source of infection into a herd where the disease does not exist or is present only in a modified form.

Or, again, animals may be introduced into an infected, perhaps apparently healthy herd, as at Kasongo, only to die of trypanosomiasis.

For example, Kivu cattle brought to the banks of the Congo; Lower Congo cattle taken up the river to Eala. Broden (3) reports that healthy cattle brought to Galiema from Dolo, about six miles away, became infected with trypanosomes. *Glossina palpalis* and big game occur about both places; the possibilities of infection are equal at both. He suggests that the Dolo cattle, which had no immunity against the Galiema trypanosome, may have been immune to another or perhaps merely a less virulent trypanosome occurring at Dolo. We are in entire agreement with him.

The cattle from Kasongo which we had sent to Lokandu (page 245) were in splendid condition. It seemed probable that they were more or less immune to the Kasongo trypanosome and would live for many months. Out of six animals sent, two were dead and two obviously ill within nine months of their arrival at Lokandu.

If for any reason, e.g., breeding purposes, it is absolutely necessary to move one or two head of cattle, they should be carefully examined for a considerable period (months) before commencing their journey. While travelling every precaution should be taken to preserve them from fly-bites. On arrival at their destination they should be isolated and not drafted into any existing herd until a second examination of some months' duration has been completed.

D. Special care must be taken in the choice and maintenance of posts for cattle-raising in areas where trypanosomiasis is liable to occur.

In the parts of the Congo visited, cattle did best in open plain country, where tsetse flies (*Glossina palpalis*) and big game (antelopes and buffaloes) were rare and pasturage good.

As Bruce first showed, big game are often infected with trypanosomes; their wholesale destruction in the neighbourhood of cattle has therefore been frequently advised. To rid a herd of other sources of infection, the infected animals existing in it must also be destroyed.

Nothing is said here concerning treatment. The results presented in another paper (page 275) are, however, quite promising enough to cause our method of treatment to be given a careful trial in trypanosome affections.

Since the blood of infected animals remains infective for some time after their death, and since some flies (*Stomoxys* and *Glossina*[1]) capable of transmitting the disease, will feed on dead animals, great care should be taken to prevent flies from reaching any part of animals slaughtered because of trypanosomiasis.[8] The *Glossinae* haunt thick bush or grass situated near collections of water; cattle should be kept in clear areas.[1] *Stomoxys* breeds in dung; the kraals should be kept scrupulously clean.

E. Game, as well as native African cattle and other domestic animals, may sometimes acquire a tolerance of certain strains of trypanosomes.

The African big game is usually said to be immune to trypanosomes. Tolerant would perhaps be a better word. A buffalo, zebra and antelopes inoculated with Nagana in Europe have died of the disease.[9] This "immunity," therefore, does not extend to every member of these species, and must be acquired by individuals. Adult, apparently healthy antelopes (and other wild animals) which are infected with trypanosomes are frequently killed. Because of the percentage* of animals infected and the abundance of biting flies, it is certain that a young antelope is infected with trypanosomes for the first time very soon after it is born, and that it will continue to be frequently re-infected during the whole of its life. The parasites so frequently found in large adult wild animals, therefore, represent either a new, recent re-infection or an original infection of long standing. In either case the animal has grown up normally, in spite of trypanosome infections; *it is tolerant of the parasites.* It seems probable that the course of trypanosomiasis in African domestic animals may some times be of a similar type.

At Kasongo trypanosomes were seen in 5 per cent. of cattle. The percentage actually infected is certainly much larger; yet this herd is increasing. There were no deaths in it from disease during our stay at Kasongo, and the cattle were in splendid condition.

Trypanosomiasis is common and often fatal among Gambian horses (6). But two horses in the Gambia found to be infected with *Trypanosoma dimorphon* in

* Bruce found 22·9 per cent. infected in South Africa. In the Congo two *Tragelaphus scriptus* (eight examined) were the only animals infected among 22 large mammals examined. It must be remembered that all these animals were examined by methods admittedly fallacious. The percentage of wild animals infected must therefore be considered to be much higher.

December, 1902, were still alive, apparently healthy, and were constantly worked in November, 1906.*

The first part of the history of Horse VI is another instance of tolerance of trypanosome infection.

As will be gathered from the notes on the animals present at various posts in the Congo, individual cattle, horses, &c., seem to be resistant, since they live for some years in places where the disease is of a very severe type. Cattle are susceptible to inoculation with *Trypanosoma dimorphon*, yet at many places in the Gambia, at Cape St. Mary for example, large herds of cattle in splendid condition graze over the same ground as the horses. (Captain Todd found trypanosomes in Gambia cattle, and he states that it is a not infrequent cause of death.)

From all these facts it is not unreasonable to suppose that, like the game, domesticated animals may thrive in spite of actual trypanosome infection and exposure to constant re-infection.

It was a "tolerance" of this sort which Schilling had in mind when he spoke of a "symbiosis" of trypanosomes with their mammalian hosts (18). He describes cattle which have apparently recovered from infection with *Trypanosoma brucei*. His later publications (19), we believe, simply confirm his former opinion expressed in (18).

Malaria in African natives forms a striking parallel to this conception. Children suffer severely from the disease.† Those who survive have parasites constantly in their blood until they become adult, and even then they are not infrequently infected, and may have short attacks of fever.

An animal long infected with trypanosomes may have short attacks of fever, with the appearance of numerous parasites in the blood, and afterwards completely recover its former tolerant status (Horse VI). From the fact that adult antelopes are sometimes heavily infected with trypanosomes, it seems likely that there may be recrudescences of the disease in the same way in them. From the conditions obtaining in the Gambia, where "sleeping sickness" has been endemic for many years, it seems probable that man may acquire a similar tolerance of *Trypanosoma gambiense* ([1], page 23). Almost nothing is known of the causes which may make the latent form of infection in animal or man virulent, and so produce the death of the individual host or an epidemic of trypanosomiasis in places where it was formerly sporadic ([10], page 319), ([5], page 27). Such an effect might conceivably be produced by (*a*) a diminution in the resistance of the host, or by (*b*) an increase in the virulence of the parasite. (*a*) Epidemics of sleeping sickness have seemed to follow hard times produced by wars and famines.[5, 10] In the individual

* One of these animals has been treated at intervals with arsenic in the form of Fowler's solution. In March, 1907, he was still in excellent condition.

† In this connection it is noteworthy that the mortality from trypanosomiasis (?) seems particularly great among calves; see Eala, Nouvelle Anvers, Tshofa, &c.

animal, hard work, lack of food and concomitant disease sometimes seem to lead to a great increase in the number of trypanosomes present and (apparently) in their pathogenic effect.

(*b*) Laboratory work has also shown that the virulence of trypanosomes propagated by direct inoculation from animal to animal may vary very greatly indeed.

Thomas and Breinl (7, pages 14 and 21) obtained a very virulent strain of *Trypanosoma gambiense*. The animal reactions of the common strain of *Trypanosoma brucei* are no longer the same as those originally described by Bruce (10, page 140). The strain of *Trypanosoma dimorphon* employed by Thomas and Breinl finally became much more virulent for laboratory animals than it was at first.

The strains of trypanosomes obtained from rats in whom *Trypanosoma brucei* had recurred after treatment by atoxyl were repeatedly found to be more virulent than the control strains (page 276).

Whether natural variations in virulence are of the same type as in these laboratory examples is unknown. An observation bearing on this point is that there was no difference between the reactions either at the first or succeeding passages of animals infected with *Trypanosoma gambiense* in the Gambia, where sleeping sickness is sporadic, and those infected at Leopoldville or in Uganda, where that disease is epidemic (11, 12).

XI. CONCLUSIONS

A. Cattle trypanosomiasis is very widely distributed in the Congo Free State.

B. *Trypanosoma dimorphon* is probably the usual infecting parasite.

C. All cattle transport should be interrupted or severely controlled.

D. The posts for cattle raising must be carefully chosen and maintained.

E. Domestic animals probably acquire a relative immunity to some strains of trypanosomes, and may even recover spontaneously (see Section IX).

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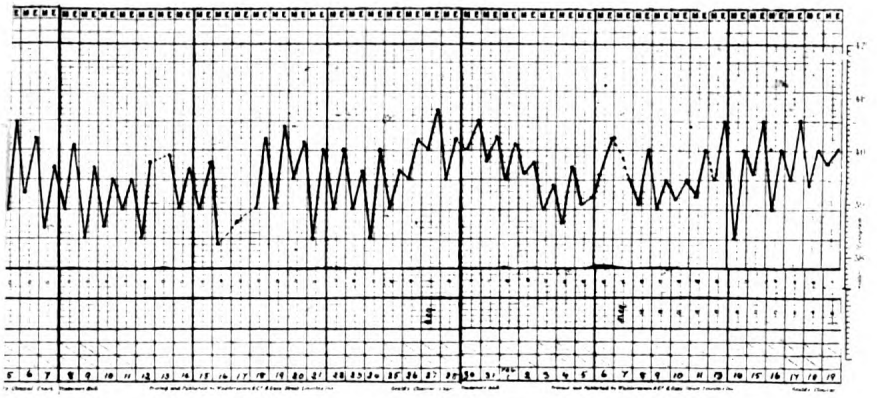


CHART I

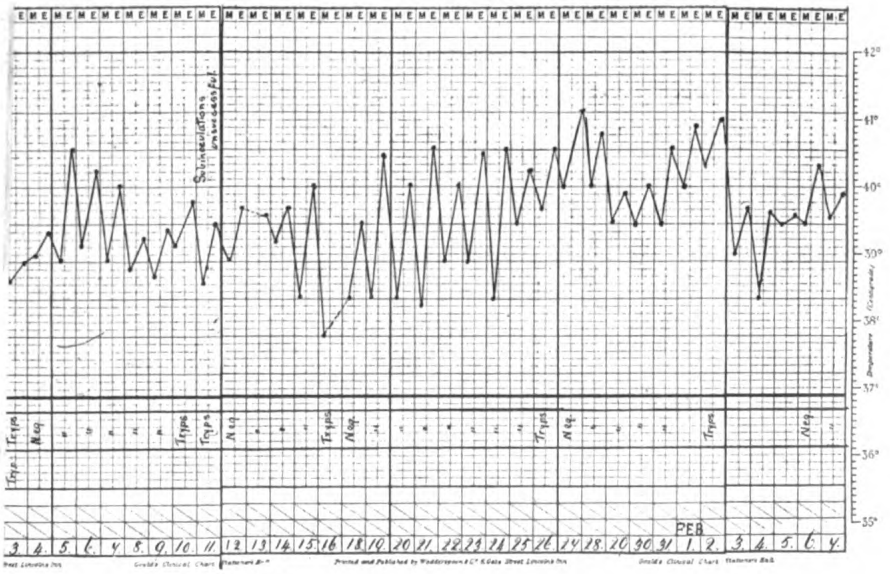


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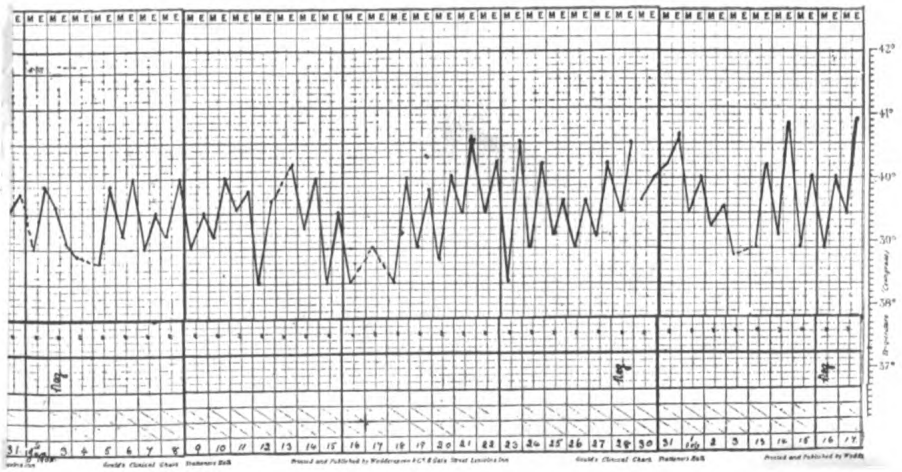
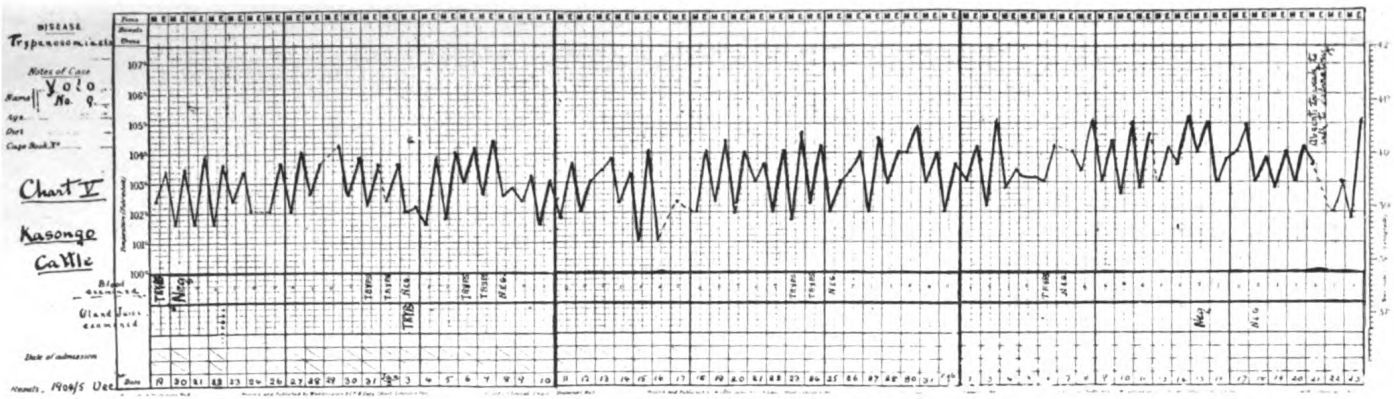


CHART III


DISEASE
Trypanosomiasis
Notes of Case
Name Yolo
No. 9
Chart V
Kasonge
Cattle
Blood examined
Gland Juice examined
Date of admission
Result

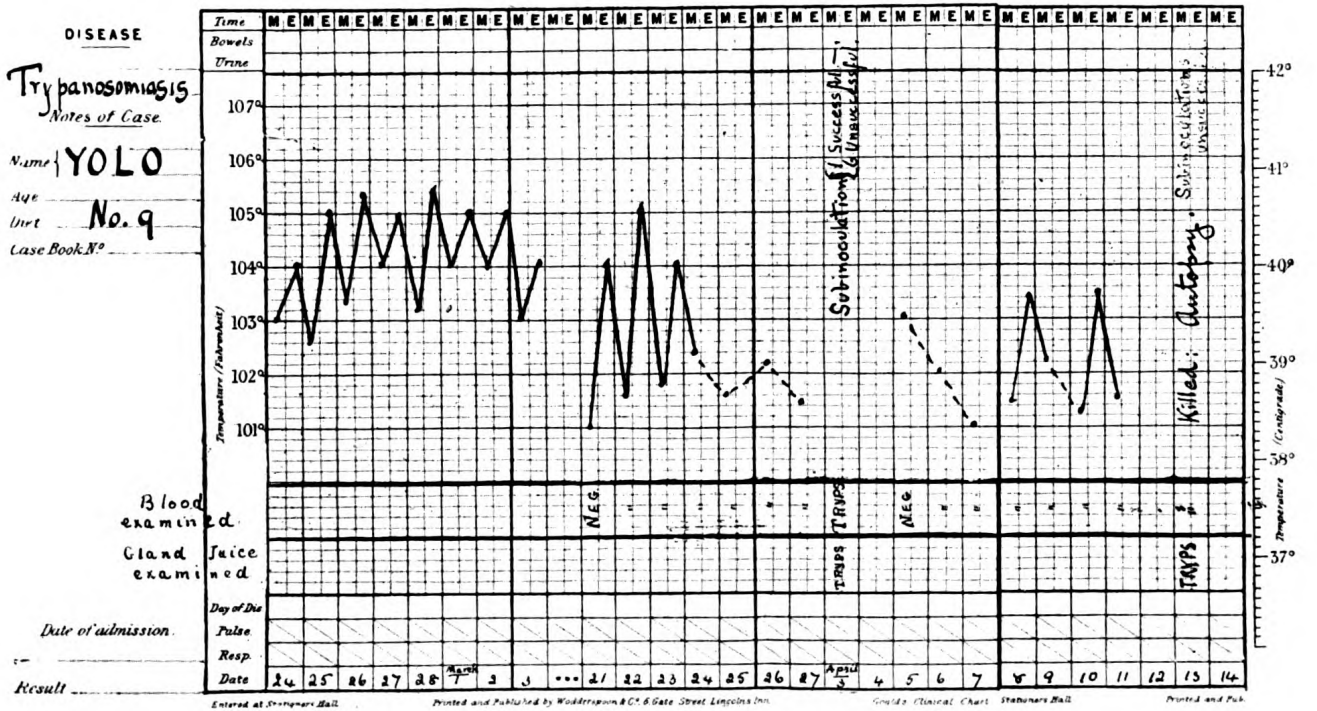

DISEASE
Trypanosomiasis
Notes of Case.
Name YOLO
Age
Diet No. 9
Case Book N°
Blood examined.
Gland Juice examined.
Date of admission.
Result
Time
Bowels
Urine
Temperature (Fahrenheit)
107°
106°
105°
104°
103°
102°
101°
42°
41°
40°
39°
38°
37°
Temperature (Centigrade)
Day of Dis
Pulse
Resp.
Date
NEG.
TRYPS
NEG
TRYPS
Killed: Autopsy.
Entered at Stationers Hall.
Printed and Published by Wodderspoon & Co. 6 Gate Street Lincolns Inn.
Gould's Clinical Chart. Stationers Hall.
Printed and Pub.

CHART V

CONCERNING THE TREATMENT OF EXPERIMENTAL TRYPANOSOMIASIS

CONCERNING THE TREATMENT OF EXPERIMENTAL TRYPANOSOMIASIS

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The present communication presents some of the results of an experimental study on the therapeutics of trypanosomiasis carried on in Liverpool.

I. PROCEDURE

Trypanosoma brucei was used in most of the experiments given here because of its great virulence for experimental animals. Results are thus obtained much more rapidly and conclusions may be drawn with greater certainty than is the case with work done with other species of pathogenic trypanosomes.

Where the comparative value of two drugs was in question, care was taken to employ animals of as nearly as possible the same body weight; the rats used in the following experiments weighed from 140 to 175 grammes. All rats were inoculated subcutaneously with approximately ·4 c.cm. of blood taken from a rat on the second day of infection and mixed with sodium citrate solution. In comparative experiments, as far as possible, all animals were inoculated with the same infective material.

The strains of *T. brucei* employed killed rats in from five to seven days after inoculation. In the later experiments a particularly virulent strain was used which killed rats in from three to four days. This strain was obtained from a rat in which trypanosomes had reappeared after treatment by atoxyl. A similar result was obtained in four out of eight observations; more work is being done on this point.

In all cases the rule laid down by Thomas and Breinl[1] was followed —that the experimental treatment of animals infected with trypanosomiasis must not be undertaken unless the disease is well advanced— that is, unless the parasites are constantly present and the animal shows obvious signs of illness (loss of weight, &c.). The poisonous dose of each drug, for the animals employed, was always ascertained as a preliminary step to experiment. The largest possible therapeutic dose was employed in every instance. The drugs were given subcutaneously in sterile solutions.

Atoxyl, unless it was otherwise stated, was given in a 5 per cent. solution in sterile water. It was found necessary to use only fresh solutions, since atoxyl in solutions deteriorates rapidly.[3]

When experiments were first commenced, the solutions of atoxyl were kept for some little time.* It was noticed that their toxic effect on animals infected with *T. brucei* was evidently great. Out of 121 rats, 40 died within 24 hours; out of 58 rats at a similar stage of infection treated with freshly made solutions of atoxyl only 10 died. This increased toxicity on standing was confirmed by observations on a solution of 5 per cent. atoxyl in water which had been kept exposed to light for seven months. Rats inoculated with ·5 c.cm. of this solution died within four hours. It is interesting to note that a similar solution of atoxyl kept for the same length of time in a dark-coloured bottle in a cupboard was not nearly so toxic.

For purposes of comparison it is well to state here that as a general rule *T. brucei* reappeared in the circulation of rats, treated by atoxyl alone, in from 16 to 25 days after the interruption of the treatment. Out of 113 rats treated by atoxyl in the same manner as was employed in our mercury experiments (see below) only three are still alive (at 126, 92 and 74 days after the cessation of treatment). In every case

* Laveran[4] has also noted the increased toxic effect of aqueous solutions of atoxyl which had stood for some time.

in the following experiments, where sub-inoculations were made from experimental animals to test whether they were infected with trypanosomiasis, from 1 to 2 c.cm. of blood were employed.

The routine examination of the blood was made in fresh $\frac{3}{4}$-inch square coverslip preparations. Occasionally the blood of important animals was centrifugalised.

For the first ten days or a fortnight after the completion of a treatment the animals were examined daily. As the animals lived longer the examinations became less and less frequent, until they were done approximately weekly. The atoxyl-treated controls were at first examined daily. When it was seen that the trypanosomes usually reappeared at about 16 to 25 days after the cessation of treatment examinations were omitted for the first two days after treatment. The blood of any animal evidently ill was of course immediately examined and, if necessary, sub-inoculations were made.

II. STRYCHNINE

Van Campenhout advised* that strychnine sulphate should be used in connection with atoxyl in the treatment of human trypanosomiasis. The following experiments were undertaken with the object of ascertaining whether strychnine sulphate has any action on trypanosomes when given alone, in conjunction with atoxyl, or when combined with arsenic or other drugs. Unless it is otherwise stated *T. brucei* was used in every case.

(1) *Strychnine sulphate alone*

(*a*) *Prophylactic.*—A rat was given 9·2 c.cm. of a 1 per cent. solution of strychnine sulphate in eight doses between October 23rd and November 7th. It was then inoculated with *T. brucei*. The infection took the usual course and the animal died on the seventh day after inoculation.

(*b*) *Curative.*—The treatment of four rats commenced on the first day of the appearance of parasites in the blood. Two rats got altogether ·3 c.cm. of a ·2 per cent. solution of strychnine sulphate. The remaining two rats received ·8 c.cm. of the same solution in two doses. The rat receiving the largest dose showed signs of strychnine poisoning but recovered. All the animals died on the third day after showing trypanosomes.

* In private communications, reports in the lay press and, latterly, in the Bulletin de l'Academie royale de medicine de Belgique (*2*). In the latter publication Van Campenhout makes it clear that he considers that the effect of strychnine is probably not on the parasite, but that it exercises a beneficial effect on the nervous tissue injured by the trypanosomes.

Two rats infected with *T. gambiense* each received 3·2 c.cm. of a ·2 per cent. solution of strychnine sulphate in five doses between the 5th and 16th November. Parasites appeared on the 26th October. Both animals died on the 23rd November.

In none of the above experiments were any signs of degeneration observed in the trypanosomes during treatment.

(2) *Strychnine sulphate followed by atoxyl*

Curative.—Two rats each received ·3 c.cm. of a ·2 per cent. solution of strychnine sulphate on the second day of showing trypanosomes in the blood. On the third day, when the animals were moribund, they each received ·7 c.cm. of a ·5 per cent. solution of atoxyl. One died. The second recovered and in the following week received three ·5 c.cm. doses of a ·2 per cent. solution of strychnine sulphate. It showed trypanosomes on the 17th day after their disappearance as a result of the atoxyl treatment, and the animal died three days later.

(3) *Strychnine sulphate given in conjunction with atoxyl*

Curative.—The treatment of two rats was commenced on the second day of the appearance of trypanosomes in the blood. From the 8th to the 17th of October each received 3·2 c.cm. of a ·5 per cent. solution of atoxyl in five doses and 9 c.cm. of a ·2 per cent. solution of strychnine sulphate in nine doses. On the 16th November (17 days after stopping the atoxyl treatment) trypanosomes reappeared. One animal died four days later. The treatment was repeated in the other (a rat subinoculated from this animal at this time died in the usual way from nagana in seven days), but trypanosomes eventually reappeared and the animal died on the 6th December.

The treatment of two rats was commenced on the third day of the appearance of *T. gambiense*. From the 26th of October to the 23rd of November each animal received 7·7 c.cm. of atoxyl in eight doses and 18·2 c.cm. of strychnine sulphate in twenty doses. One of the rats died on the 2nd December, the other on February 26th, 1907; each had many trypanosomes in its blood.

(4) *Strychnine sulphate preceded by atoxyl*

Curative.—(*a*) The treatment of two rats was commenced on the first day of the appearance of *T. orucei* in the blood. ·7 c.cm. of a ·5 per cent. solution of atoxyl was given in two doses, and then, on the seventh day after the trypanosomes had appeared, treatment by strychnine sulphate was commenced. For a fortnight the animals received daily 2 c.cm. of a ·2 per cent. solution. The parasites reappeared on the seventeenth day after the atoxyl was stopped, and both animals died of trypanosome infection.

(*b*) Four rats each received ·5 c.cm. of atoxyl on the third and fifth days of appearance of *T. brucei* in the blood. Two of them had no further treatment; trypanosomes appeared and both died within 22 days. The two remaining rats each received 2·2 c.cm. of strychnine solution in four doses on the sixth and tenth days of infection. One died of trypanosomiasis in 17 days, the other in 23 days.

(5) *Strychnine arseniate*

Curative.—Eight rats inoculated with *T. brucei* were given from ·5 to 1·5 c.cm. of a ·06 per cent. solution of strychnine arseniate on the second day of the appearance of trypanosomes in the blood.

The course of the disease was entirely uninfluenced and the animals died on the third and fourth day from the appearance of the trypanosomes. In some of the experiments signs of degeneration of the parasites were noticed.

(6) *Strychnine arseniate followed by quinine arseniate*

Curative.—Four rats infected with *T. brucei* each received 2·5 c.cm. of a ·06 solution of strychnine arseniate and 2·5 c.cm. of a ·1 per cent. solution of quinine arseniate, as in the preceding experiment. The drugs were given separately, at twelve hour intervals, each in three doses on three successive days. The animals died on the fourth day, that is two days after the appearance of parasites in the blood. A few degenerate trypanosomes were noticed on the second day.

From these experiments we conclude that

Strychnine has probably no harmful effect on *Trypanosoma brucei* or *T. gambiense*.

III. ATOXYL, FOLLOWED BY MERCURY

Our work has been directed by the idea[5] that perhaps the recurrences in cases of trypanosome infection, after treatment by atoxyl, might be due to some resistant stage of the parasite which survived the first treatment by atoxyl and later gave rise to recurrences of trypanosomes more or less refractory to atoxyl treatment. It was thought that these hypothetical resistant forms might be influenced by some other drug. A series of experiments on the treatment of animals infected by trypanosomes was therefore commenced in which atoxyl was followed by some other drug. Up to the present the best results have been obtained by the use of atoxyl followed by mercury. We present the results of this treatment on rats infected with *Trypanosoma brucei*.

(*a*) *Atoxyl followed by the bichloride of mercury*

The same solution of atoxyl was used as in the preceding experiments. The mercury was given subcutaneously in a 0·1 per cent. solution (Liquor Hydrargyri perchloridi, B.P.). The mercury was never given until parasites could no longer be found by the examination of coverslip preparations. That they were probably usually absent from the blood in an infective form is shown by the fact that none of four animals inoculated, as controls, with blood taken from these experimental animals after their treatment by atoxyl have ever become infected.

EXPERIMENT (110).—Four rats received subcutaneously 0·5 c.cm. of a 5 per cent. solution of atoxyl (the same strength of solution is used in the same way unless otherwise stated, in all the following experiments) on the third day of infection, the eighth day after inoculation, with trypanosomes. On the four following days 2 c.cm. of the 0·1 per cent. solution of perchloride of mercury was given subcutaneously (the same solution was employed in the following experiments).

Trypanosomes were never afterwards seen in any of these rats. One rat died 42 days after inoculation with trypanosomes. Another, being moribund, was killed 95 days after inoculation. Trypanosomes were not seen at the autopsy on either animal, and a rat sub-inoculated from the rat killed has never shown trypanosomes. The remaining two rats are still alive, 181 days after inoculation. Rats and mice sub-inoculated from them on the 92nd and 156th days have never shown trypanosomes and are still alive.

EXPERIMENT (134).—Six rats received 0·8 c.cm. of atoxyl solution in two doses on the second and third days after inoculation. The infection was severe; one rat died before, two others just after the first dose of atoxyl. Two of the remaining rats received no further treatment. One died of trypanosomiasis in 27 days, the other in 22 days, after the cessation of treatment.

In the last rat the course of atoxyl was followed by 1·5 c.cm. of mercury perchloride given in two doses on the fifth and eleventh days after inoculation. Trypanosomes never reappeared. Thirty-one days after inoculation this rat seemed ill; it was therefore killed and another rat was sub-inoculated. Trypanosomes never appeared, and the sub-inoculated rat died of skin disease three months later.

EXPERIMENT (149).—Five rats each received 0·5 c.cm. of atoxyl on the third day after inoculation. Two rats received no further treatment; one died (cause of death ?) in 16 days after inoculation, the other of trypanosomiasis, in 28 days (parasites reappeared three days before death).

In three rats the atoxyl was followed by 2·7 c.cm. of mercury perchloride given in four doses on the sixth, seventh, tenth and eleventh days after inoculation. One died (cause of death ?) in 40 days. The remaining two are still alive, 155 days after inoculation. Mice sub-inoculated from them 130 days after inoculation have not become infected.

EXPERIMENT (159).—Six rats each received 0·5 c.cm. of atoxyl on the fourth day after inoculation. Three rats which received no further treatment showed trypanosomes in from 12 to 24 days and died in from 15 to 27 days after inoculation.

The other three rats received 2·7 c.cm. of perchloride in four doses on the seventh to tenth days after inoculation. Trypanosomes reappeared in none of them. One rat died on the eighth day after inoculation (cause ?), another on the eleventh day (pneumonia). The last rat is alive 133 days after inoculation, and mice sub-inoculated from it on the 108th day are not infected.

EXPERIMENT (161).—Two rats each received 0·5 c.cm. of atoxyl on the fifth day after inoculation. One rat had no further treatment and died three days later.

The other rat received 1·9 c.cm. of perchloride in three doses on the eighth, tenth and twelfth days after inoculation. *Trypanosomes reappeared* on the 15th day. Another 0·5 c.cm. of atoxyl was given at once, but the animal died next day.

Treatment was probably commenced too late here; both animals were almost moribund when the atoxyl was given. It is possible that the result might have been better in the mercury-treated rat, had a larger dose of atoxyl been given. One dose is probably insufficient for so heavily-infected an animal. It may be questioned whether the parasites were ever entirely absent from the circulation.

EXPERIMENT (284).—Six rats were inoculated; one died on the third day. On the fourth day the remaining five rats received 1 c.cm. of atoxyl in two doses. Two had no further treatment. Trypanosomes reappeared in them in 20 to 23 days after the cessation of treatment, and they died a couple of days later, of trypanosomiasis (30 days after inoculation).

Three rats, after the atoxyl, each received 2·7 c.cm. of perchloride in four doses on the fifth to eighth days after inoculation. Trypanosomes were not again seen in any of them. One rat died (spleen enlarged, cause of death ?) 20 days after inoculation; the remaining two are alive 123 days after inoculation. Mice inoculated from them 98 days after inoculation were not infected and are still alive.

EXPERIMENT (294).—The treatment of six rats was commenced on the fourth day after inoculation; their infection was heavy and two died on the fifth day. The remaining four rats received 1·4 c.cm. of atoxyl in three doses on the fourth, eighth and tenth days after inoculation. Two rats had no further treatment; trypanosomes reappeared in them in 34 and 41 days after the cessation of treatment. They died two to four days later (53 days after inoculation).

In two rats atoxyl was supplemented by 2·7 c.cm. of perchloride given in four doses on the fifteenth to nineteenth days after inoculation. Trypanosomes have reappeared in neither of them. On the thirtieth day after inoculation one of these rats died (cause of death ?); after the second dose of atoxyl this animal had shown very marked signs of intoxication. The remaining animal is still alive 95 days after inoculation. A rat and mouse sub-inoculated from it 43 and 70 days after inoculation are alive and uninfected.

EXPERIMENT (296).—Four out of 12 rats died during the night of the second day after inoculation. On the third and fourth days the remaining eight received 1 c.cm. of atoxyl in two doses. Four rats received no further treatment; one of them died almost immediately. Parasites reappeared in the other three in from 10 to 41 days after the cessation of treatment and all died in from 16 to 49 days after inoculation.

On the fourth, eighth, tenth and eleventh days after inoculation 2·7 c.cm. of perchloride was given in four doses to each of the remaining four rats; one died (*intoxication* ?) after the first dose. All of the three rats were negative until 36 days after inoculation, when *trypanosomes reappeared in one of them*. The other two are still negative at 84 days after inoculation; mice sub-inoculated from them at 59 days after inoculation have not shown trypanosomes. The rat in which trypanosomes reappeared received 1 c.cm. of atoxyl in two doses on the day its relapse was detected, and the second day after. It then received 1·7 c.cm. of perchloride in three doses on the fifth, seventh and ninth days after the relapse. Parasites again reappeared in 14 days after the relapse. Atoxyl was again immediately given (1 c.cm. in two doses on successive days); the trypanosomes disappeared as usual from the peripheral circulation. The animal died unexpectedly seven days later. Trypanosomes had not reappeared in its peripheral blood; spleen and lymphatic glands were enlarged.

This is the only instance of a recurrence of trypanosomes after a satisfactory combined treatment by atoxyl and mercury. In this experiment treatment was certainly commenced very late. There was a smaller interval between the administration of the two drugs than is usual; but there were probably no infective trypanosomes left in the peripheral circulation after the atoxyl treatment, since a rat subinoculated at that time has not shown trypanosomes and still lives.

EXPERIMENT (297).—On the third and fourth day after inoculation 10 rats received 1 c.cm. of atoxyl in two doses; two rats died during the night of the third day. Four rats received no further treatment. Trypanosomes reappeared in them in from 13 to 24 days after the cessation of treatment and they died two or three days later.

The remaining four rats received 2·7 c.cm. of perchloride in four doses on the sixth to tenth days after inoculation; trypanosomes were not again seen in any of them. One rat died (cause ?) 25 days after inoculation. The remaining three are still alive, 56 days after inoculation, and mice sub-inoculated from them on the thirty-first day are alive and have never shown trypanosomes.

When viewed in the severest light the results of these experiments are distinctly encouraging. The combined treatment by atoxyl and mercury was given to 25 rats in advanced trypanosomiasis; all would certainly have died a very few hours later. Of them 13 are still living (after 181 to 56 days) while all the controls, which received, at the same time, the same quantity of atoxyl alone, are long since dead. Experiments have been commenced in which the effect of repeated doses of atoxyl is compared with this combined treatment by atoxyl and mercury.

The three rats in Experiments 159, 161 (see above) which died soon after the commencement of treatment, should probably be disregarded, since in these instances treatment was commenced very late. The cause of death of many of the treated animals is obscure. It must be asked whether death was ever wholly due to intoxication from the drugs administered. There were two recurrences of trypanosomes (Experiments 161 and 296). Two subinoculations in rats made from the treated animals dead in Experiments 110 and 134 are still alive. It therefore seems doubly doubtful whether death was in these instances due to trypanosomiasis. If these facts are considered our results become still more encouraging.*

(*b*) *Atoxyl followed by Donovan's solution*†

EXPERIMENT (310).—Eight rats received 1 c.cm. of a 3 per cent. solution of atoxyl in two doses on the fourth and fifth days after inoculation. Two rats died during the fourth night. On the sixth, eighth and tenth days 1·5 c.cm. of Donovan's solution was given in three doses to the remaining six rats. All of them are still alive 44 days after inoculation; mice sub-inoculated on the nineteenth day are alive and have not shown trypanosomes.

This experiment quite confirms those which have preceded it; it seems certain that such a combined treatment as we describe should be given a thorough trial in the treatment of human and animal trypanosomiasis.

* Experiments of a similar type, at present under observation, on *T. brucei* and *T. gambiense* in guinea pigs, rabbits, dogs and donkeys have so far been in complete accord with the results obtained in rats.

† 1 per cent Iodide of mercury, 1 per cent. of Iodide of arsenic in water.

It has been abundantly proved by the work of many previous observers and by our own preliminary experiments that mercury perchloride alone will not cure trypanosomiasis. That it has a marked beneficial effect when preceded by atoxyl is shown by the results communicated in this paper. How has the mercury acted? A tempting explanation would be that atoxyl attacks and kills the usual active well-known phase of the trypanosome found in the peripheral circulation, but that there exists somewhere in the tissues an inert,* resting phase, or that such a phase is formed from the active phase by the action of the atoxyl.[12] On this phase the atoxyl has no poisonous action or an ineffectual one, but on it the mercury salt, which conversely has no action on the active form, has a powerful poisonous action. The result is that the inert form exists after atoxyl treatment in the tissues. On stopping the drug, or on the trypanosome becoming inured to it,† there is a recurrence of the active form and a fresh outbreak of the disease. The mercury prevents recurrence by killing the inert form.

This hypothesis suggests the application successively of two or more remedies, each attacking a different phase of the existence of protozoan parasites.

Two such remedies attacking two successive phases are likely to prove more successful than two remedies attacking the same phase.‡ For example, Mesnil and Nicolle and Aubert[9] have shown that atoxyl in alternation with their aniline colours (Ph and Cl) is practically not more effectual than atoxyl alone.§ Here two agents identical in action are administered; the novelty of a mercury salt after atoxyl lies in the fact that the mercury is not active in the same phase as the atoxyl.

From this point of view it would be of interest to discover whether the mercury salt would be equally effectual after the aniline trypanocides, and this we are proceeding to investigate.

* We owe this suggestion primarily to Dr. Thomas.

† Ehrlich (11) has produced strains of trypanosomes uninfluenced by atoxyl.

‡ Trypanocidal substances known to have a decided action on the active stage of trypanosomes are the following:—by Ehrlich and Shiga (7), trypan-red; by Ehrlich (8), trypan-blue; by Mesnil and Nicolle (9), colours Ph (synonym of trypan-blue) and Cl; by Wendelstadt and Fulmer (10), malachite green; by Thomas and Breinl (1), atoxyl.

§ Combined treatment along these lines has been attempted by many authors, notably by Laveran (6) and Thomas and Breinl (1).

It is concluded that :—

(1) In the treatment of rats infected by *Trypanosoma brucei* the administration of atoxyl followed by mercury perchloride gives better results than does uncontinued treatment by atoxyl alone.

(2) The combined treatment should be given a careful trial in natural trypanosome infections of man and animals.

(3) The treatment must be commenced as early in the infection as possible; full therapeutic doses of the drug must be given; fresh solutions of atoxyl must be employed;* probably the mercury should not be given until the parasites have been driven from the peripheral circulation.

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* By medical men, who will take the necessary precautions, atoxyl is best given intravenously (1).

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1905	Hudson, Charles Tilson
1905	Illington, Edmund Moritz
1906	Jeffreys, Herbert Castelman
1907	Kennan, Richard Henry
1907	Kenrick, William Hamilton
1904	Khan, Saiduzzafor
1904	Laurie, Robert
1907	Le Fanu, George Ernest Hugh
1905	Macfarlane, Robert Maxwell
1906	Mackenzie, Donald Francis
1904	Maclurkin, Alfred Robert
1905	Maddock, Edward Cecil Gordon
1907	Maddox, Ralph Henry
1907	McCarthy, John McDonald
1904	McConnell, Robert Ernest
1905	Moore, James Jackson
1904	Nicholson, James Edward
1905	Nightingale, Samuel Shore
1906	Pailthorpe, Mary Elizabeth
1906	Palmer, Harold Thornbury
1906	Pearse, Albert
1904	Philipson, Nicholas
1905	Radcliffe, Percy Alexander Hurst
1907	Ryan, Joseph Charles
1906	Sampey, Alexander William
1904	Sharman, Eric Harding
1906	Smithson, Arthur Ernest
1906	Taylor, Joseph van Someron
1906	Taylor, William Irwin
1904	Thomson, Frank Wyville
1906	Tynan, Edward Joseph
1907	Vallance, Hugh
1904	Walker, George Francis Clegg
1906	Watson, Cecil Francis
1906	Willcocks, Roger Durant
1906	Williamson, George Alexander
1905	Young, John Cameron

EDITORIAL NOTICE

By order of the Committee of the Incorporated Liverpool School of Tropical Medicine, the series of the Reports of the School, which have been issued since 1899, will be followed, from January 1, 1907, by the Annals of Tropical Medicine and Parasitology, of which this is the third number.

The Annals are issued by the Committee of the School, and will contain all such matter as was formerly printed in the Reports—that is to say, accounts of the various expeditions of the School and of the scientific work done in its laboratories at the University of Liverpool and at Runcorn. Altogether twenty-one Memoirs, besides other works, have been published by the School since 1899, and of these ten, containing 519 quarto or octavo pages and 95 plates and figures, were published during the two years 1904 and 1905; and there is no reason to suppose that this rate of production by the workers of the School alone will diminish in the future. In addition, however, to School work, original articles from outside on any subject connected with Tropical Medicine or Hygiene may be published if found suitable (see notice on back of cover); so that, in all probability, not less than four numbers of the Annals will be issued annually. Each number will be brought out when material sufficient for it has been accumulated.

CONCERNING CERTAIN PARASITIC PROTOZOA OBSERVED IN AFRICA

CONCERNING CERTAIN PARASITIC PROTOZOA OBSERVED IN AFRICA*

Being the Eighth Interim Report of the Expedition of the Liverpool School of Tropical Medicine to the Congo, 1903-5

BY THE LATE
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PART II†

MAMMALS (continued)

DOGS

Leucocytozoön.

In films of fresh blood taken from an apparently healthy native puppy in the Gambia free gregarine-like forms were seen which moved slowly in much the same way as do free drepanidia. Intracorpuscular forms were not seen; the stained slides have been mislaid.

From this observation it seems probable that parasites resembling the haemogregarines described in Indian dogs‡ may also occur in the Gambia.

*We desire to acknowledge our indebtedness to Dr. J. W. B. Hanington for his kind aid in the preparation of the drawings accompanying the present communication, and in the examination of some of the slides of Arthropoda.

†Part I of this paper appeared in Memoir XXI of the Liverpool School of Tropical Medicine.

‡For example, Christophers: *Leucocytozoön canis*, Scientific Memoirs of the Officers of the Medical and Sanitary Department of the Government of India. New series, No. 26, 1905.

BIRDS

Very many birds of widely different species were examined. Only those in whose blood parasitic protozoa were found are mentioned here; several trypanosomes found in Gambian birds are described in a previous publication, Memoir XI of this School.

Proteosoma was never seen. *Halteridium* was very common among the small birds and pigeons examined in the Gambia. In the Congo *Halteridium* was seen only once or twice in small birds and in none of those in which the parasites described below were found.

ASTURINULA MONOGRAMMICA. (A grey hawk common in the Congo Free State.)

One of these birds was shot about midday at Lokandu and its blood was at once examined. In fresh specimens actively moving trypanosomes were seen; dried films were therefore made at once. On studying them parasites of the same type as the *Leucocytozoön* of Danielewsky[1] (Plates XX-XXIII, figs. 48, 72) were seen to be present in large numbers.

A second hawk of this species was shot at Lusambo. Its blood contained a trypanosome of apparently the same species as that observed in the first hawk; the *Leucocytozoön* was not seen (one small slide examined).

Leucocytozoön ziemanni.

Under the name of *Leucocytozoön ziemanni*, Lühe[2] gives a good review of our knowledge concerning this parasite; to his list of references two additional papers[3, 8] should be added, in which the occurrence of similar parasites is reported in America and in Egypt. Although the name *Leucocytozoön* has the priority, its use seems unfortunate, since the parasite to which it here refers is probably parasitic in other cells besides white cells[4, 6] and it has no resemblance to the recently-described, undoubted leucocytozoa of mammals.

Male (figs. 48-61) and female (figs. 62-67) gametocytes of the types fully and finally described by Schaudinn[4] and previously reported by Sacharoff, Ziemann,[5] Laveran[6] and Berestneff[7] were frequent. In addition, many other undescribed forms of the parasite were seen. From the nature and scantiness (four slides) of our material, it is

impossible to attempt a far-reaching study of the parasite. Nevertheless, a fairly complete description is given of some forms constantly observed. This is done with the purpose of insisting on what an excellent object for study this type of parasite is; especially since some of the forms seen seem in some measure to harmonise with Schaudinn's description[4] of the development of *Spirochæta ziemanni* in the blood stream.

Schaudinn gives no particulars but simply states that the development of *Spirochæta ziemanni* differs in no important point from that of *Hæmoproteus noctuæ*.

In our specimens the forms already frequently described as makrogametocytes (figs. 66, 67, 69, 72, 73), and mikrogametocytes (figs. 48, 49) are easily recognisable, and in the main their appearance is in accord with previous descriptions. It is noticeable that in all forms of the parasite the nucleus of the host-cell is but little flattened, and practically never takes the dumb-bell-like shape described by former observers in similar parasites. The position of the host nucleus in the adult parasite varies according to the position in which the parasite dries on the slide. Although it is usually placed laterally at a point about midway between the two extremities of the parasite (fig. 49), it may occur in any position. It has frequently been extruded (fig. 65), and gametocytes without any included host-cell nucleus are not uncommon (fig. 66).

The general structure of the parasites of each sex at all stages of development is the same. Each parasite has periplast, ectoplasm, endoplasm and nuclear bodies. The "periplast" consists of a non-granular, pink-staining (always ?) sheath which invests the whole of the parasite. In it run fairly numerous faint, non-staining, longitudinal, or, if the parasite is twisted, apparently diagonal striations (these can be seen by careful examination in even the youngest parasites). The periplast with its striations is, as a rule, best seen in the more lightly staining mikrogametocytes (fig. 50). In the adult forms the periplast is prolonged at either extremity into long slender processes. When the matured parasite becomes rounded, preparatory to conjugation, the periplastic sheath (fig. 77) is thrown off together with the effete host-cell nucleus (fig. 31).

A similar process probably occurs in the change from the resting to the motile stages (Schaudinn)[4].

Appearances in young parasites (Plate XX), as well as in the

gametocytes, whose effilated extremities have a distinctly granular structure, indicate that an "ectoplasm" exists quite distinct from the periplast. In the young parasite its tendency is to stain blue; in the adult it is pink.

The "endoplasm" is the most striking part of the whole parasite. So much so that it, with the nuclear bodies, has been described by some as the whole of the parasite. In the makrogametocytes the endoplasm stains a deep blue (fig. 73). It frequently contains large numbers (20-30) of coarse, or fine, vividly chromatophilic granules and many small, circular non-staining areas—"vacuoles." The endoplasm of the mikrogametocytes (fig. 49) stains much less deeply, and rarely contains granules or vacuoles. The endoplasm appears to consist of a system of wide alveoli filled with a more or less granular protoplasm. This structure is particularly well seen in the makrogametocytes. The sexual variation in the endoplasm exists, though in a minor degree, in some of the smallest parasites (endoplasm measures 4μ by 3μ).

The form of the "nuclear bodies" varies enormously, according to the sex and stage of development of the parasite.

The amount of chromatophilic material—"nuclear material" in its widest sense—present is much greater in the adult male (fig. 53) than in the female parasites (fig. 67). For convenience of description the various nuclear structures observed at different stages of development are given the following names: "chlamydoplasm,"* "nucleus," "blepharoplast," with an accompanying "vacuole," "the line" and certain "dots," whose nature is certainly problematical. It must be clearly understood that these names are only descriptive and that our material has not enabled us to definitely ascertain the functions of the bodies to which they are applied. The fixation and staining employed are both faulty. The parasites are often distorted by pressure of surrounding red cells and partially destroyed—occasionally useful details can be gleaned from such "fortuitously dissected" specimens (figs. 11, 12, 15, 36). Romanowsky is, unfortunately, a diffuse stain, and the details of dense structures, such as blepharoplasts and the "line," are often hopelessly obscured.

* From Gr. χλάμυς = a cloak. This name is adopted as a descriptive term to indicate the diffusely pink area surrounding the nucleus. It is employed in preference to other terms indicating similar structures since we know nothing, and wish to assert nothing, concerning its function.

The " chlamydoplasm " is a diffuse, light-purple-staining area whose outlines, though very irregular, are, nevertheless, usually definite. In the female parasites (figs. 66, 67, 72, 73) and in males with lines (figs. 33-46) it is comparatively compact. In older mikrogametocytes its mass becomes greatly increased (figs. 50-56) and its outline is less definite (figs. 58, 61). It may lie compactly about the nucleus or stretched out (fig. 76) and distributed in any way through the endoplasm. If it is divided, one part remains with the nucleus, the other is about, or near, the blepharoplast (Fig. 50).

The " nucleus " is employed as an indefinite term to designate the denser collection of chromatin enclosed in the chlamydoplasm. Its shape varies considerably; possibly its nature does so also.

The " blepharoplast " is the term applied to a very deeply staining collection of chromatin usually occurring quite alone in the endoplasm, and showing a distinctly granular structure (figs. 3, 55, 48). In the younger parasites (figs. 3, 5) only two to four granules, arranged in pairs, can be counted. In the older parasites (figs. 68, 71) the blepharoplast consists of one central granule and from six to eight other granules joined together by fine lines and situated about the periphery of the sphere of translucent chromatophilic material in which they are placed, and outside the chlamydoplasm. As a rule, no blepharoplast-like body exists separate from the nucleus in parasites which show the line formation; rarely one is present (fig. 37). In such parasites the line seems to have split at the end opposite to that at which the free blepharoplast is placed. This " blepharoplast " is probably formed by division of the " nuclear " mass of chromatin. An oval non-staining or more lightly staining area, the " vacuole," is very constantly associated with the blepharoplast in parasites of all ages. It may vary greatly in size (figs. 53, 73). " Dots," or chromatic granules similar to those illustrated in figs. 15, 19, 23, 47, 59, were seen in a few instances. They occur in parasites of all sexes and sizes in any position. It is impossible to say anything concerning their function from our specimens. In one parasite of the type illustrated in fig. 71 a chain of five pairs of granules, arranged in a column, occurred within the chlamydoplasm and in immediate connection with the nucleus. In the same way a column of three pairs of granules has been seen placed beside the blepharoplast. In one or two cases a pair of granules was joined by a fine thread.

Unfortunately, our material does not permit a full description and explanation of the developmental phases of these nuclear structures. We can only describe a few of the appearances observed. One process, however, the formation of a "line," seems common to all sexes and ages of the parasite. The line is widest about the middle, and tapers to either extremity. It is usually gently curved; rarely it seems to be slightly wavy. This effect is probably produced by the over-lying striations of the ectoplasm.

We have not determined exactly how this line is produced, nor what is its significance. The morphological changes attending its appearance in the younger parasites are as follows:—The nuclear material, in the youngest parasites (fig. 2, 6), consists of a deeply chromatophilic area, connected with which is a larger pinkish area often containing a few chromatophilic granules. A line is projected apparently from the denser nuclear mass (fig. 1). At the distal end of this line a smaller, densely-staining area appears (fig. 3). The steps intervening between this stage and that shown in figs. 16, 17, 18, where a commencing line is apparently arising from the lighter, not the darker, part of the chromatic material, have not been determined. It is possible that the forms shown in figs. 4, 5, 9, 10 may intervene at this period. In slightly injured parasites, of all ages (figs. 15, 36), it is seen that the line is composed of several (up to four counted) filaments. Sometimes (figs. 28, 33, 33a) it may split longitudinally, apparently normally; and (?) in such cases the nucleus sometimes also divides (fig. 39). Multiplication of the parasite has not been seen. This appearance is the only one observed which in any way suggests division. The line may also divide transversely in the following manner. A portion of the line situated in the chlamydoplasmic area becomes thinned (figs. 26, 32, 40, 41).* An oval pink area differentiated from the rest of the nucleus surrounds this constriction. At either side of the constriction a dark granule develops in the line (fig. 32). Connecting these granules is a very fine dark line or "axial filament." (The "nucleus" in one instance (fig. 26) was connected to one of these granules by a line.)

In another, slightly squashed specimen a filament connected the line with a differentiated "nuclear" area situated in the chlamydo-

* Occasionally the blepharoplast or nucleus may lie over the line and so, through a defect in staining, produce an appearance resembling a constriction.

plasm and then with a "blepharoplast." In some cases the granules and connecting filament can be discerned in the line even before the constriction becomes visible; in two or three specimens a wavy blue line was associated with the "line" (figs. 38, 46, 62). We make no suggestion concerning its nature. The further development of this process was seen in only one specimen (fig. 44), here the two halves of the line are widely separated. The proximal extremity of each is capped by a dark granule and surrounded by diffuse chromatin material, while between them runs the faint axial filament, whose apparent origin has been described.

The process as thus observed seems comparable to the first stages of the formation of motile apparatus in the trypanosome-like stage of *Halteridium (Hæmoproteus) noctuæ*, Schaudinn. The possibility suggests itself that the formation and transverse division of the "line" may represent the third division of nuclear material described by Schaudinn, and that the axial filament may represent the flagellar apparatus of a future trypanosome. Unfortunately, our specimens permit us to go no further than merely to suggest this hypothesis.

Since line formation occurs in parasites of all sizes, it is a process common to all ages of the parasite. This is also true of the development of the trypanosome-like, motile stages of *Hæmoproteus*, and is another point of resemblance between these two processes.

Objections to this interpretation of the line formation are:— (1) None of the smallest trypanosome-like or spirochaete-like stages representing the motile stage of the youngest intracorpuscular parasites were seen. This is strange in a bird so heavily infected. The trypanosome which was present (described below (figs. 29 and 30)), if it has any connection with the leucocytozoön, probably represents the motile stage of a mikrogametocyte.

(2) Forms (figs. 45, 46, 63, 64) in which the length of the line has become much extended do not seem to harmonize with this hypothesis. Advanced stages of transverse division of the line were seen but rarely (figs. 26 and 44 are unique).

The line may lie in any position. It may be connected with its nucleus at its centre or by one end (figs. 45, 74). Although it is usually almost totally in the endoplasm of the parasite, its extremities may extend into the ectoplasm (figs. 39, 46, 74). According to the way in which the parasite has dried on the slide the line may be at

the side of, above, or below the host-cell nucleus. The line is the most resistent part of the parasite, and is often intact when the rest of the parasite has been destroyed in the making of the film.

Young forms, smaller than those illustrated (figs. 6, 7, 8, 13), occur in which it is impossible to detect any structure beyond the presence of a spot of chromatin in a slightly larger area of blue endoplasm. Every intermediate stage occurs between these tiny organisms and such larger, line-bearing parasites as figs. 24, 25. Intermediate stages, without the line, are not seen between parasites of this size and the adult gametocytes. Intermediate stages with the line are frequent (fig. 26). The younger parasites are spherical or have rounded ends (figs. 4, 13, 25); as they become larger their extremities become pointed and affiliated. Many figures in Plate I indicate that the younger parasites are amoeboid. This, and the fact that one or two very young parasites were seen free in the plasm, suggests that the parasites may be able to wander from host-cell to host-cell without the development of a motor apparatus. Such a process is described in *Hæmoproteus noctuæ*.[4] It is evident from changes in form of the adult parasites (figs. 59, 63), apparently not due to artefacts, that they retain up to a late stage something of the plasticity of the younger parasites. The younger parasites seem to apply themselves to (figs. 1, 4, 25) or to enter (figs. 13, 16) the host-cells. The larger parasites engulf them (fig. 26).

It is noteworthy that the parasite's endoplasm is always in close connection with the cell nucleus. As far as staining reaction and appearance goes, it is evident (figs. 6-8, 13, 21, 25) that the host-cells often are mononuclear white cells; very rarely granular leucocytes are attached.

Probably as the infection becomes older, the host-cell, particularly the nucleus, becomes larger and stains deeply (figs. 24, 26, &c). It is noteworthy that the nuclei of the host-cells harbouring female parasites are more often extruded and when present are more degenerated than is the case in the male parasites (figs. 62, 73). Since Romanowsky is a stain which does not penetrate, host-cell nuclei lying beneath the parasite are frequently almost unstained. In such specimens it is often difficult to make out the relative arrangement of host-cell and parasite.

Adult Female forms.

Adult female parasites, makrogametocytes, are rather less numerous and larger than adult male parasites. Their general characteristics, as outlined above, are: deeply-staining endoplasm, containing more or less numerous chromatophilic granules and small clear spaces—"vacuoles"; and small amount of nuclear material, as compared with the male parasites. The average measurements of the most usual type of makrogametocytes (figs. 65, 67) are:—Total length, 55·6 μ; endoplasm, length 20·3 μ, breadth 9·8 μ. Slender forms (figs. 62, 73) in which these measurements were 63·1 μ, 22·5 μ, and 8·5 μ respectively, as well as stumpier forms (fig. 69) measuring 47 μ, 16 μ, and 14 μ, were constant types. Much effilated forms (fig. 72) are rare. They occur most frequently in the thickest parts of our blood smears.

In the most usually seen form (fig. 67) the nuclear material forms a diffusely-pinkish area (chlamydoplasm ?), usually without a sharply-defined limit, but possessing a definite oval contour. In it, or immediately adjacent to it, is the more or less deeply-staining, often granular, blepharoplast (?). Adjoining the blepharoplast an oval, more lightly stained area—the vacuole—can usually be distinguished (fig. 73). Other less conspicuous, dense, chromatic areas may also occur in the chlamydoplasm (fig. 66). The phenomena attending line formation in the female (figs. 62, 63, 64) seem to be analogous to those described in the male and in the young forms; because of the darkly staining endoplasm it is, however, very difficult to follow them.

No changes were observed in the nuclei of the female cells which seem peculiar to them. For this reason no special description is given of them; the few drawings reproduced indicate the resemblance between the nuclear changes in the two sexes. The coarse alveolar structure of the endoplasm, common to all forms of the parasite, is particularly defined in the makrogametocytes. The curved outlines of the alveoli, merging into one another, often give the appearance of wavy blue lines running through the parasite. The granules vary greatly in number and size. There may be as many as fifty. Some are almost dust-like; usually they are larger, and may measure almost ·5 μ in diameter. They frequently occur in pairs and seem to be placed superficially in the parasite. They often lie in lines along the faint striations of the ectoplasm. Of the origin and nature of

these granules we can say nothing certain. Granules of similar appearance sometimes occur in much smaller numbers in mikrogametocytes.

A few rounded forms (fig. 31) are present in which the host-cell nucleus has been extruded and the ectoplasm thrown off. These are possibly parasites prepared for fertilization.[6] Nothing was ascertained concerning the nuclear changes at this stage.* Such parasites measure about 14μ by 9μ.

Adult whet-stone-shaped parasites occur (fig. 70) which possess general characters intermediate between the male and female adult types described. They are, therefore, not readily referable to either sex; but our material does not permit an assertion as to whether an indifferent form exists or not.

Adult Male forms.

Besides the forms in which the line occurs or is developing (fig. 34), there are other parasites where the nuclear material follows a development of different type (figs. 48, 52); it is impossible to say definitely whether there is any connection between these two processes.

Apparently an early stage of this second process is illustrated in figs. 48, 70, 71, 75. There is more or less diffuse and abundant chlamydoplasm. In the chlamydoplasm occurs a denser, at first circular or spherical, mass of chromatin—the "nucleus." (It may rarely be placed just outside the chlamydoplasm.) Outside of the chlamydoplasm, but occasionally in connection with a detached portion of it, is a second denser mass of chromatin—the "blepharoplast." The blepharoplast is identified by its granular nature and by its darker staining. The nuclear material becomes arranged in a thick semicircular arc (figs. 49, 76.) At its centre usually occurs a dot, often connected with the extremities of the arc by fine lines (figs. 53, 55). The arc of chromatin becomes hemispherical and the dot increases in size (figs. 50, 52) until the place of the nucleus is taken by two irregularly oval chromatin masses of approximately equal size (fig. 51). A stage preliminary to this process is possibly illustrated in fig. 47, where four brownish granules, of quite a different colour from the chromatin granules of the blepharoplast, occur in close connection

* The line in the nucleus of fig. 31 makes it very doubtful to what stage this parasite really belongs.

with the nucleus. This specimen is unique; somewhat similar granules once occurred (fig. 59) in connection with the blepharoplast.

It is possible that the division of the nucleus may be by mitosis. Some half-dozen forms like figs. 54, 56, 59 were seen. Unfortunately, it was never possible to distinguish the individual spindle fibres or to count the individual chromosomes. Some spindles, however, had a distinctly fibrillar appearance, and the chromatin was usually very distinctly granular at this stage; in one specimen (fig. 56) the granules could almost be counted—there seemed to be from seven to nine. The further development of this process was not observed.

The type of parasite in fig. 68 is probably an early stage of this second process. The nuclear concentration in the chlamydoplasm is indistinct and circular. The blepharoplast is very well marked; its thread-connected granules are distinct, and not infrequently a chromatophilic granule occurs well outside the blepharoplast, but still connected to it by a well-defined reddish thread (fig. 68). (In one such parasite the blepharoplast had divided into two equal parts connected by a thread.) This apparent extrusion of granules from the blepharoplast occurs, but less frequently, at other stages (figs. 49, 50).

Figs. 51 and 60 are larger parasites apparently undergoing a similar process. In fig. 57 the blepharoplast seems to be extruding two granules, while the nucleus is commencing to divide. In fig. 60 the nucleus, now almost outside the chlamydoplasm, is almost completely divided, while the granules in the blepharoplast have become much more distinct. Fig. 58 is probably a stage in this process. It is suggested that this second process is possibly concerned with the prostages of mikrogamete formation.

A trypanosome.

Six trypanosomes which cannot be identified easily with any parasite already described were seen in the slides containing the leucocytozoön. All were of approximately the same type. The usual measurements were about I,* 3·7μ; II, 10·5μ; III, 1·8μ; IV, 21·4μ;

* The measurements of trypanosomes in this communication were made according to the formula described by us on page 88 of Memoir XXI of this School.
I—Posterior extremity of the parasite to centre of the blepharoplast.
II—From the centre of the blepharoplast to the posterior border of the nucleus.
III—From the posterior to the anterior border of the nucleus.
IV—Anterior border of nucleus to posterior extremity of the body of the parasite.
V—Length of free flagellum.
VI—Breadth of body at its widest part.
The total length of the parasite is also given as a measurement.

V, 11·8μ; VI, 3·7μ. Total length, 49μ. A perfect flagellum was seen only in one parasite (fig. 29). In fig. 30 the flagellum is much shorter; the greater breadth of this parasite is probably due to its being slightly flattened.

The blepharoplast is a very darkly-staining oblong lying longitudinally in the parasite and placed just posterior to an ill-marked vacuole. It obviously consists of a collection of four or more granules. The flagellum, after forming the thickened edge of a wide (1·5μ), ample, lilac-staining undulating membrane, ends in one instance (fig. 30) in a small carmine-coloured, possibly bi-lobed, expansion, in immediate apposition to the blepharoplast. The granular, palely staining nucleus measures about 2 by 1·3μ, and lies in a sharply-defined palely-staining area (3 by 2·5μ), situated rather posterior to the middle of the body of the parasite. In one instance a dark karyosome-like granule, lying in the clear space, is placed close to the nucleus. The nucleus is obscured by the striations of the body, and it is seen with great difficulty. The finely-alveolar body cytoplasm is striated through almost its whole length. The much-pointed posterior extremity (especially in fig. 29) is very lightly stained. Here, as in the finely-drawn-out, darkly-stained anterior extremity of the body, striation can not be detected. At the level of the nucleus about eight, more or less, light striations (myonemes ?) can be distinguished running longitudinally. Ordinarily (fig. 29) the striae are placed at equal distances. In fig. 30 they are so arranged as to make the cytoplasm appear to be arranged in dark striae disposed in pairs.* By careful examination of fig. 30, it was thought that five pairs could be distinguished.

The irregular clear areas in the cytoplasm, as illustrated (figs. 29, 30), occur in the majority of the parasites. They are not thought to be artefacts. They seem to be non-staining, refractile granules rather than vacuoles.

The periplast does not stain as pinkly as is usual in trypanosomes, but it can, nevertheless, be distinguished as a clear refractile envelope about the body of the parasite.

* Compare with the myonemes in the motile forms of the mikrogametocytes of *Spirochæta ziemanni*.

"BUSH FOWL"

A few trypanosomes, measuring about 50μ in length, were seen in a red-legged bush fowl at Tshofa. Stained preparations were not made. The parasite was remarkable for the extraordinary length and the flagellum-like fineness of its posterior extremity. About one quarter of the total body length lay posterior to the blepharoplast.

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BYCANISTES BUCCINATOR (the trumpeter hornbill).

*Trypanosomes.**

Two distinct trypanosomes, one small and the other large, were seen in a bird of this species shot near Coquilhatville in the Congo Free State. The smaller trypanosomes (Plate XXIV, figs. 1, 2, 3, 4, 5, 6) were by far the most numerous. Their appearance in stained specimens varies very considerably, so that it is possible to describe three distinct types, although forms intermediate between them can be distinguished. These may be called the "slender" (figs. 1, 2), "broad" (figs. 3, 4), and "stumpy" (figs. 5, 6) forms, in accordance with their general appearance. To some extent the structure of these forms varies with their dimensions. The stouter forms usually stain more lightly, have the looser cytoplasm, have no vacuole at the posterior extremity, and their large nuclei do not extend completely across their bodies.

This association of qualities is not constant, however, and all gradations were seen between forms possessing them and the, as a rule, darkly-staining and more compact slender forms. All these forms are, therefore, considered to be merely variations of one parasite, which may be described as follows:—

The blepharoplast stains very densely and more darkly than either nucleus or flagellum. Its position may vary from the extreme posterior extremity, in particular in the "broad" and "stumpy" forms, to a spot 1μ or more from the end of the parasite (fig. 1). It is oblong in shape and is seen to be granular; in several specimens at least four granules, sometimes arranged in pairs, can be counted (fig. 6). Although the arrangement of these granules occasionally suggests commencing division, none of the ordinary longitudinal division forms were seen. The blepharoplast may be placed longitudinally, obliquely, or transversely in the parasite. Just anterior to it there is often a well-defined vacuole, or, when that is absent, a more lightly stained area.

The nucleus frequently extends completely across the body of the parasite and is almost always surrounded by an area stained more lightly than is the remainder of the body. The relation between the size of the nucleus and the type of parasite varies considerably;

* The description of this parasite is reproduced by permission from the Journal of Medical Research. Vol. XVI, No. 5, March, 1907.

as a rule, in the "broad" and "stumpy" forms the nucleus seems relatively larger, of looser texture, and stains more lightly. Chromatic granules, number undetermined, occur in the nucleus of each type of trypanosome (fig. 1).

In several preparations (figs. 3, 6) two small darkly-stained and closely-apposed chromatophilic granules, surrounded by a pinkish area, occur in close connection with the nucleus, but just outside the nuclear membrane. In one instance a well-marked chain of oblong, twin chromatophilic granules runs forward, in an unstained area, from the anterior edge of the vacuole for about a quarter of the distance between the blepharoplast and the nucleus. This structure recalls a similar appearance observed in *Trypanosoma karyozeukton.**

Whether staining deeply or lightly, the cytoplasm of the small trypanosomes is always granular, but the fineness of the granules varies greatly. Well-marked striations of the protoplasm occur in each form, although they are best seen in the broader forms (figs. 3, 4, 5). The striations are evidently a superficial structure, and are usually wound spirally about the parasite. In some specimens a single striation can be followed for almost two complete turns (figs. 4, 5). From their arrangement it is very difficult to estimate the number of striations, but these seem to be about 7 to 8 (figs. 4, 5).

The whole parasite is enclosed in a pink-staining periplast, which can be plainly seen at the posterior extremity and in some parasites all along the edge of the body, where it is unobscured by the undulating membrane (fig. 3). The ample undulating membrane seems relatively widest in the "slender" forms.

The usual dimensions of each form are given in the following table:—

USUAL MEASUREMENTS OF THE SMALL TRYPANOSOME*

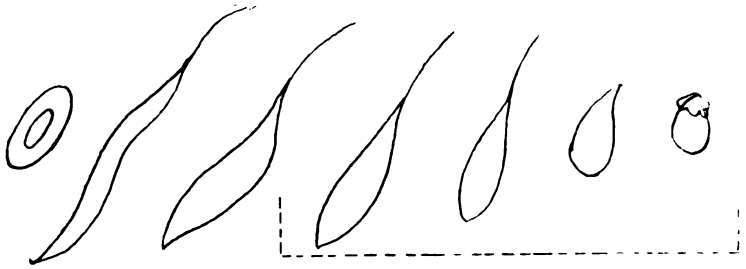
			STUMPY FORM		BROAD FORM		SLENDER FORM
Measurement	I	...	1·0 μ	...	1·6 μ	...	1·6 μ
,,	II	...	7·0 μ	...	8·0 μ	...	10·4 μ
,,	III	...	2·0 μ	...	2·4 μ	...	2·0 μ
,,	IV	...	7·3 μ	...	10·4 μ	...	7·2 μ
,,	V	...	7·8 μ	...	8·0 μ	...	9·6 μ
Breadth	VI	...	7·0 μ	...	4·8 μ	...	2·8 μ
Total length	VII	...	25·1 μ	...	30·4 μ	...	30·8 μ

* See page 297

* Dutton and Todd, 1903. Memoir XI, Liverpool School of Tropical Medicine, page 53.

Only two examples of the large trypanosome (total length about 64μ), were obtained. Unfortunately, both are so obscured by surrounding red cells that it is impossible to reproduce them. Their blepharoplast is placed much nearer to the nucleus than to the posterior extremity (and in one instance in a vacuole). The nucleus almost extends across the body. The undulating membrane is ample and the flagellum seems to be comparatively short. The body of the parasite is striated longitudinally; at the level of the nucleus only seven striations could be counted. In one parasite an appearance resembling a longitudinal striation was present in the undulating membrane. The dimensions of this type are: posterior extremity to centre of blepharoplast, 28·3μ (in one parasite only 17μ); centre of blepharoplast to posterior border of nucleus, 3·3μ; anterior border of nucleus to termination of body, 18·3μ; free flagellum, 8·3μ; width 5·8μ.

At Coquilhatville lack of time prevented a careful examination of the parasite in fresh preparations. Some months later another hornbill was shot, and its blood was found to contain trypanosomes resembling the small type described above. Fresh coverslip preparations of blood were kept at room temperature (28° C.) and watched for some hours by Dr. Inge Heiberg. The changes in form indicated in the following diagram were observed to occur in three hours.



Two hours after the preparation was made normal parasites were still seen. Occasionally pairs occurred joined by their flagella. After three hours both normal and very short "stumpy" parasites were seen, while after four hours, longitudinally dividing, spherical and irregular, certainly degenerating, parasites were present.

Eighteen hours after the preparation was made dividing forms, similar to those seen at the fourth hour were still present, while a stouter trypanosome than those seen at the commencement of the observation now appeared for the first time.

REPTILES

A considerable number of snakes, chameleons, lizards, monitors and tortoises of several varieties, as well as three or four crocodiles, were examined in both the Gambia and in the Congo. Parasites were found only in the instances described below.

SNAKES

Trypanosomes.

In stained smears of blood taken from a puff-adder caught in the Gambia, a single trypanosome-like object was seen.

The measurements are as follows:—

Measurements: *	I	... 6·0 μ
	II	... 6·4 μ
	III	... 2·4 μ
	IV	... 21·6 μ
	V	... 8·4 μ
	VI	... 0·6 μ
Total length of parasite		45·0 μ

The body is long, narrow and tapers at each end to a pointed extremity. The body stains a light blue except for two areas between the blepharoplast and the nucleus, which stain dark blue. The first dark area, extending from the blepharoplast anteriorly, is 14μ long; the second, following a short interval, is 3μ long. The nucleus is composed of red chromatin granules and occupies the entire width of the body. The flagellum is a very slender filament. Only a suggestion of an undulating membrane can be seen.

Drepanidia. †

Drepanidia of an ordinary type were frequently seen in the same snake. The nuclei of the cells containing them were displaced, but

* See page 297.

† In the present state of our knowledge, it seems futile to attempt the identification of a Hæmogregarine from its "drepanidium-like" form alone. For a review of what is known concerning the haemogregarines of amphibians and reptiles, see Lühe; in Mense's "Handbuch der Tropenkrankheiten," page 206. Barth, Leipzig.

not injured; neither was there any destruction of the cell cytoplasm. Two drepanidia were often seen occupying one blood corpuscle, sometimes on either side of the nucleus, and sometimes at one end of the corpuscle, which was then double the usual length (Plate XXV, fig. 52). Two forms of drepanidia were seen, one with a finely-granular, striated, blue-staining cytoplasm (Plate XXV, figs. 50, 55), and the other with a lighter-staining, more loosely-woven cytoplasm, which contained vacuoles, of irregular size and number, at both ends (Plate XXV, fig. 51). These latter forms were often much shorter and wider than the others. The drepanidia measured from 11 to 17μ in length, and from 4·5 to 8μ in width. Free forms of the usual type were seen; multiplication forms did not occur. Folded, "two-shanked," intra-corpuscular parasites were not seen.

A few of the curious bodies illustrated in Plate XXV, figs. 53, 54, and in Plate XXVIII, figs. 56-58, were seen in the blood of this snake. They were rods from 10 to 19μ long and about 1μ wide. They stained homogeneously red with Romanowsky; they were, however, a deeper red at the edges than in the middle. The ends of the rods were somewhat rounded, but never well-defined. In a free single rod there was a deep red dot near the centre (Plate XXV, fig. 53). Two rods were always found in a cell, and they were usually of about the same length, though occasionally one was longer than the other. The rods were sometimes placed parallel, sometimes crossed upon one another, and sometimes came together at one end. They usually occurred in cells which were lighter-stained and had a rounder, redder nucleus. They were sometimes, not always, found in cells containing drepanidia. In two cases free parasites were seen; in one case two rods were found on a degenerated drepanidium; in the other case, a single rod was found free.

Dr. L. Sambon, by a study of slides of blood from other snakes in which these bodies occur more frequently, has shown that they represent the curled-up remains of the capsules vacated by drepanidia.*

An Unidentified Parasite.

A single rounded parasite of the type seen in frogs (page 335) was found in the snake (Plate XXV, fig. 55). It measured 2·2μ in diameter. It contained short red rods arranged, like the spokes of a wheel, about a small central red mass.

* Private communication.

CROCODILES

Trypanosomes.

In stained smears of blood taken from a crocodile (*Crocodilus cataphractus*?) shot in the Congo, a trypanosome with a length of about 35μ (not including flagellum) was seen. The body was about 2μ wide, the blepharoplast was 3μ from the posterior extremity and the nucleus was near the centre of the body and occupied its entire width. The undulating membrane was well developed.

Drepanidia.

Drepanidia of an ordinary type were seen in the same crocodile (Plate XXV, fig. 49). They usually measured 12·5μ in length and 4·5μ in breadth. The nuclei of the host cells were displaced but not injured. No destruction of the cytoplasm of the blood corpuscles was observed, and no division forms were seen. Folded, "two-shanked," intracorpuscular parasites did not occur.

LITERATURE

MINCHIN, GRAY AND TULLOCH. Proc. Roy. Soc., Series B., Vol. 78, p. 251. Report the presence of a large trypanosome in a crocodile in Uganda. They give no further particulars.

TORTOISES

Trypanosomes.

In stained smears of blood taken from a tortoise caught in the Gambia, trypanosomes of one type were seen (Plate XXV, fig. 45). The measurements are as follows:—

Measurement	I	1·2 to 3·5μ
,,	II	7·5 to 18·5μ
,,	III	2·2 to 3·5μ
,,	IV	12·5 to 23·0μ
,,	V	10·0 to 15·0μ
,,	VI	2·0 to 3·5μ
Total length of parasite		35·0 to 58·0μ
Width of membrane		2·0μ

The body tapers gradually towards both extremities, which are sharp-pointed, the anterior extremity being more slender than the

posterior. The cytoplasm is finely granular, and in it are several round unstained spaces 1 to 1·5μ in diameter. In one specimen there were seven of these spaces anterior to the nucleus, one at the posterior edge of the nucleus and four posterior to that. The body has fine longitudinal striations. The blepharoplast consists of at least four granules embedded in a matrix, and is about 1·1μ long; a clear area is in connection with it. From it the well-developed undulating membrane extends in folds to the anterior extremity. The nucleus is round, sometimes slightly longer than wide, and occupies two-thirds of the width of the body. No division forms were seen.

*Drepanidia.**

Drepanidia of an ordinary type were seen in the same tortoise. The nucleus of the host-cell was displaced, but not injured. Two parasites were sometimes seen in one blood corpuscle. No destruction of the cytoplasm of host-cells was observed and no multiplication forms were seen. Some parasites have a cyst wall about them while they are in the corpuscles. This same encysted condition was observed in extra-corpuscular parasites. Many of the drepanidia have coarse, colourless, refractile granules (Plate XXV, fig. 47), either throughout, or only in one half of, their bodies. Two forms of parasites were observed; in one the nucleus stains a dark purple and has a dense structure, as has its cytoplasm (Plate XXV, fig. 46); in the other, which is much larger, the nucleus stains a bright carmine, and the structure of the nucleus and cytoplasm is looser (Plate XXV, fig. 48). The drepanidia measure from 10·4 to 17μ long and from 4 to 5·5μ wide. Multiplication forms were not seen. Folded, "two-shanked" intracorpuscular parasites did not occur.

AMPHIBIA

The only representatives of this class examined were frogs and toads. The blood of several hundreds of these was searched for parasites in either the Gambia or the Congo.

FROGS AND TOADS

Trypanosomes.

In the Congo *Trypanosoma loricatum* vel *costatum*[2] (Mayer, July 1843) was found in the blood of representatives of the following

* See footnote to page 303.

species of frogs and toads:—*Rana galamensis* (D. and B.), *Rana oxyrhynchus* (Sund.), *Rana mascarensis* (B.), *Rappia marmorata* (Rapp.), and *Bufo regularia* (Renn.). A considerable portion of them was infected. Blood from these infected amphibia was examined fresh and stained. In the fresh examinations, blood was carefully taken aseptically from the heart or, if it was desired to keep the frog alive, either from a leg vein or a toe. Coverslip preparations were made and examined periodically. Blood was kept in sealed capillary pipettes for varying periods and was then used for making films for staining. Blood was taken from the frogs and, with coverslip preparations already made, was examined at all hours of the day and night. Preparations from the organs and bone marrow and of fluid from the body cavities were examined; trypanosomes were seen only in the blood.*

Trypanosoma loricatum was present in almost every frog infected with trypanosomes. It was frequently associated with trypanosomes of any of the types described below. As a rule, it was noticed that parasites of the *Trypanosoma sanguinis*[4] type (Plate XXVI, fig. 27) were associated with *Trypanosoma inopinatum*-like forms and with the leaf-like forms[3] (Plate XXVI, fig. 28); *T. mega*[4] (Plate XXVII, figs. 35-39) occurred with forms resembling *T. karyozeukton*[4] (Plate XXVIII, figs. 40-42) in everything save the absence of the specific chain of granules. In 99 per cent. of the frogs infected with trypanosomes *Drepanidium* was also seen to be present. As a rule, if there were many trypanosomes there were also many drepanidia. Striking exceptions to this rule were, however, seen.†

In fresh blood *T. loricatum* is practically a frilled operculum of protoplasm, somewhat pointed at the posterior end. It is convex on one surface and concave on the other. From the median line of

* In a single paper Mayer (2) described two parasites under two specific names, *loricatum* (or *costatum*) and *rotatorium*. More recent work has shown that he was probably dealing with two forms of one trypanosome. One of these names must therefore disappear. We retain the name *loricatum* since it was originally applied to a parasite resembling that type of trypanosome which seems to be the adult form of the haematozoon under discussion. A perusal of the present paper makes it evident that various other forms of *T. loricatum* have received specific names. These must also eventually disappear; but until the life history of this parasite has been completely worked out it is scarcely worth while discussing this point.

† For descriptive purposes names already given to various types of trypanosomes are frequently used in this paper. They are used without question to designate parasites resembling those described under these names by the authors quoted.

the concave surface at a little distance from the posterior extremity arises the flagellum. The margins of the organism are roughly serrated. It moves backwards and forwards slowly, now turning over on itself and now bending antero-posteriorly. As many authors have observed, its rate of progression is very slow. It is, therefore, easily watched under the microscope for considerable periods. A side view is seen in fig. 1*, a dorsal view in fig. 2, and a cross section in fig. 3.

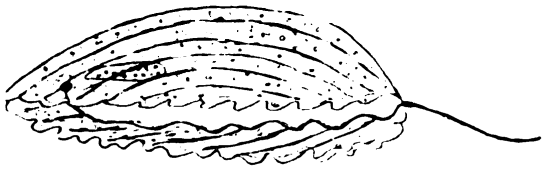


Fig. 1.

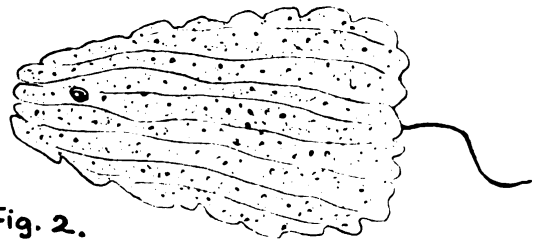


Fig. 2.



Fig. 3.

T. loricatum has a peculiar method of division, which we observed most completely in preparations of fresh blood of *Rana galamensis*, kept aseptically at 72° F. to 89° F. for two or three days.

Amongst recent authors, Franca and Athias[2] and Bouet[6] have seen parts of the cycle described in this paper. Franca and Athias[2] saw representatives of the type of *T. loricatum*, in fresh and stained specimens, become rounded and, in one case, segment several times. In stained specimens they saw a trypanosome of their *T. rotatorium* type become spherical and divide. They describe most interesting nuclear changes in which the blepharoplast seems to play the part of a centrosome, and forms suggesting mitotic division are described. In these same specimens small round parasites, possessing blepharoplast and nucleus, which may develop two flagella, occurred. A small trypanosome of their *T. rotatorium* type was also present.

* The figures in the text are drawn diagrammatically from actual living specimens. Figs. 20 and 24 are enlarged 2000 diameters, the remainder 1000 diameters.

They describe one or two anomalous forms whose position cannot be determined at present. Bouet[6] made cultures from the blood of frogs containing trypanosomes of the *T. loricatum*[2] and the *T. rotatorium*[2] types. The results obtained with either form of parasite were identical. Herpetomonas-like parasites (with an ill-developed undulating membrane, however) were the most usual forms in cultures. It is to be noted that every form of parasite between this type and the trypanosomes originally present in the frog's blood could be seen in the cultures.

Some of the parasites, directly after the preparation was made, were seen to have lost their striations at one or both ends and to have become granular. This process results in a swelling at one or both ends (fig. 4). Later the whole parasite becomes granular; during this

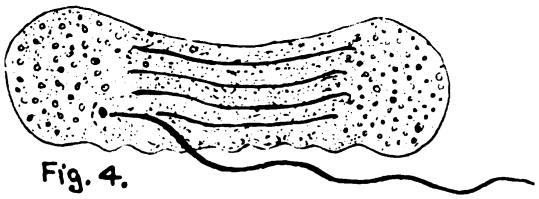


Fig. 4.

process a well-marked nucleus and blepharoplast are present. The body now becomes rounded, and the undulating membrane is gradually peeled off. The flagellum is attached to one end of the organism (fig. 5). The parasite soon becomes completely spherical (fig. 6), and in this condition its diameter is about 24μ. The flagellum is still

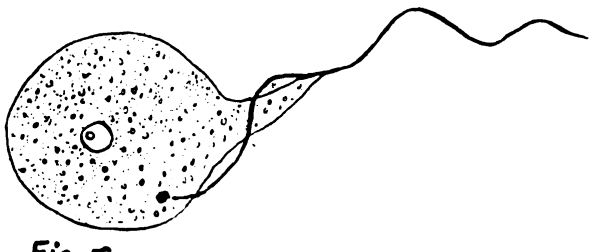


Fig. 5.

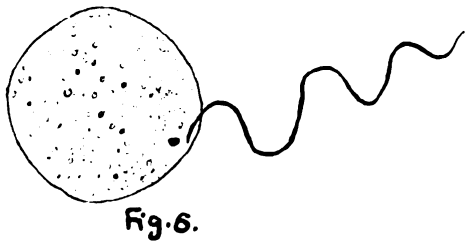


Fig. 6.

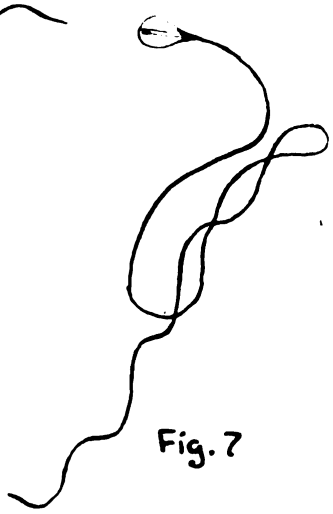


Fig. 7

attached, but soon is cast off and is seen lying in the serum, with the blepharoplast attached (fig. 7). Unfortunately, discarded flagella were never seen in stained specimens. It is, therefore, impossible to state whether the whole of the blepharoplast is cast off or not. If the whole of it is discarded a new body strongly resembling a blepharoplast is formed very quickly; because rounded parasites of all sizes from one equalling the original rounded *T. loricatum* to those only 5μ in diameter possess two chromatic areas, one the nucleus proper, the other resembling a blepharoplast.*

It is suggested by some forms that part of the blepharoplast at least may remain when the flagellum is thrown off. Chromatophilic granules not infrequently occur in the cytoplasm of trypanosomes which probably belong to this period. Concerning their nature we can say nothing.

The flagellum after being shed loses its lively movements in three or four seconds, and dies. The parasite is now round and granular with a highly refractile nucleus (fig. 8).

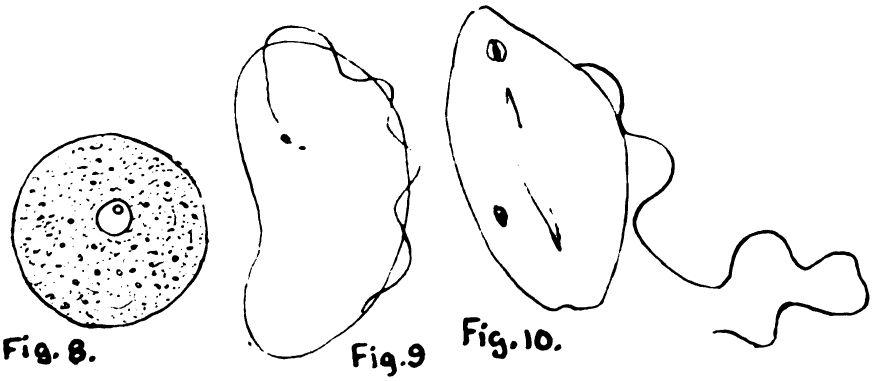


Fig. 8. Fig. 9 Fig. 10.

In one observation the parasite lost its striations, became rounded and lost its flagellum in eight minutes after the preparation was made (figs. 9, 10).†

* It would be expected (2) (8) that the blepharoplast would be newly formed from the nucleus.

† The times given for the periods occupied by the various developmental changes represent actual observations, and may be taken as approximate averages. The same changes may take place more or less rapidly; very frequently the changes take longer than has been indicated.

It then became amoeboid, and in fifteen minutes had moved across two fields (Zeiss, 1·30 aperture, achromatic objective; No. 2 eyepiece). The nucleus became distinctly visible (even before the flagellum was completely cast off), and its structure could easily be made out (fig. 11).

The parasite now elongates (fig. 12), a constriction appears, and almost before division is complete a similar constriction takes place in each of the daughter cells to form four cells, all the cells being almost of the same size as the original mother cell. The four cells divide into eight, the eight into sixteen, the sixteen into thirty-two, and the thirty-two probably into sixty-four.*

Preparatory changes were seen in the nucleus before the first division. These changes were difficult to follow owing to their rapidity. At one point a rapidly-moving little tongue process appears, surrounded by a very small differentiated area (fig. 12). In a second

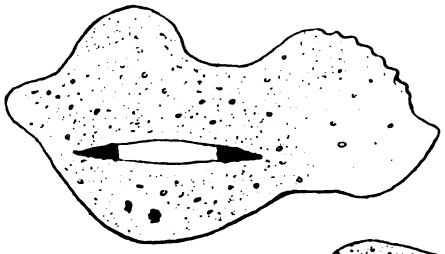


Fig. 11.

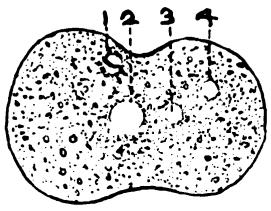


Fig. 12.

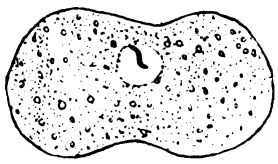


Fig. 13.

or two a dull refractile area becomes visible a little distance from it; a second later another one appears, but it is somewhat smaller, and then still another (fig. 12); but almost before this latter has formed, the four apparently merge into one large, highly-refractile, rounded area in which the little tongue is seen moving (fig. 13). This area is an obvious feature in many of the cells before division is complete, at first very bright in appearance, it gradually fades away; thus in one case it was formed at 3·21, it was much less visible at 3·24, it could not be distinguished at 3·25, and the cells had completely

* Danielewsky states that he has seen 120.

divided at 3·30. Another observation showed the area commencing to be visible at 1·55, very bright after two minutes with the little tongue plainly visible; it was not so bright at 2, and had vanished at 2·03; at this time division of the cytoplasm was well advanced. Another tongue and area was very bright at 2·07, and gone at 2·26. In one case just before its disappearance, the little halo divided, showing a delicate, thread-like connection between two dots (fig. 14). These separated completely into two (fig. 15), and then both disappeared.

A dot was sometimes discernible at one end of the "tongue." Clear areas, smaller and quite distinct from those described above, occasionally appeared for a few seconds in the cytoplasm of the trypanosomes before the first division. One of the products of the first division not infrequently divides before the other. Often one

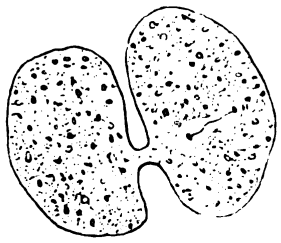


Fig. 14.

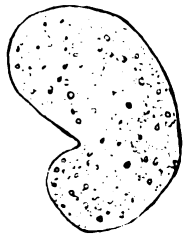


Fig. 15.

or two of the products of the third, fourth or later divisions divides no further. Indeed, the development of the parasite may be arrested at any stage. Such individuals become rounded and very granular and probably usually disintegrate. Some of them may become encysted resistant forms.[6]

In dividing, the cells rotate one upon the other in opposite directions, so that the long axis of one comes to lie at right angles to that of the other (fig. 16). The nuclei were indistinct. The time taken to turn around in one case was about thirty minutes.

In one instance the single rounded organism had divided, in five hours and forty-two minutes, into sixteen cells, which were all apparently inside the outer covering of the original trypanosome (fig. 17). In six hours and a half, many of the cells had divided

again, and in seven hours there were counted thirty-two cells which measured 8μ in diameter. In seven hours and a half, forty-one cells were counted, though there were probably more.

The next change occurring in these cells is the acquirement of a flagellum. Each becomes ovoid, then pear-shaped, and from the more rounded end a flagellum is produced (fig. 18); there are always one or two parasites in each colony which remain spherical and develop no further. The colony of cells now takes on a lively motion. After a while the young trypanosomes become free and

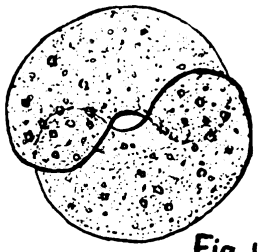


Fig. 16.

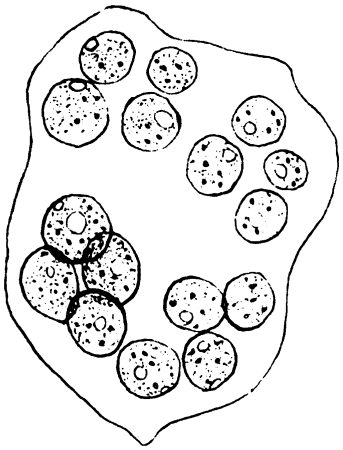


Fig. 17.

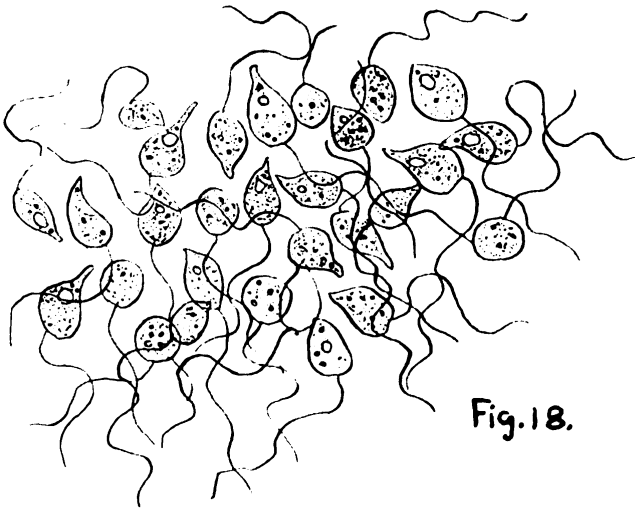


Fig. 18.

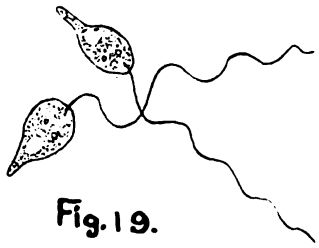


Fig. 19.

their movements increase in rapidity. They divide rapidly by splitting in their longitudinal axis, and thus in the field containing the original cells, and for many fields around are seen large numbers (10 to 15 to a field) of young actively-moving trypanosomes.

The young trypanosomes when first formed from the cell by the acquirement of a flagellum, had the shape and size (× 1,000) indicated

in fig. 19. A large or small vacuole is generally seen near the pointed end. One or two highly refractile dots occur in the cytoplasm. The movements are rapid and take place solely by the lashings of the flagellum, which acts as a tractellum. The cytoplasm does not take part in the production of locomotion even when the parasite has completely separated from its fellows. These small parasites have a herpetomonas-like form; this was especially so after division, when the body of the young parasite is drawn out and its width is almost equal throughout the whole length. No evidence of an undulating membrane was detected in these parasites; nor was the exact



Fig. 20.

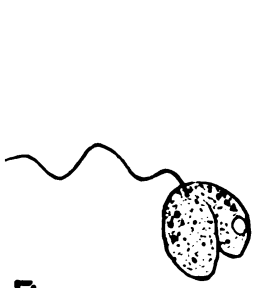


Fig. 21.

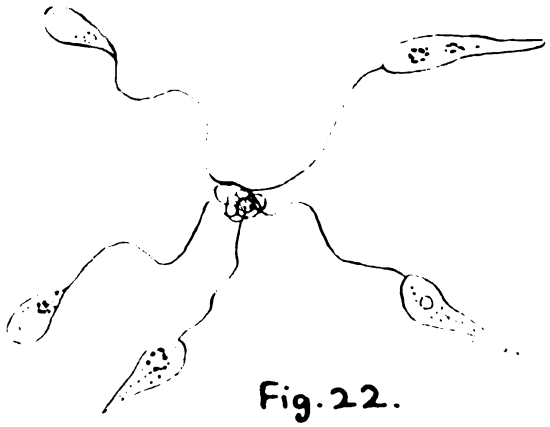


Fig. 22.

termination of the flagellum in the cytoplasm ascertained in fresh preparations. In preparations examined thirty hours and forty-five minutes after they were made, the very active trypanosomes present were all of the shape shown in fig. 20. They contained two refractile areas.

In one preparation small free parasites were seen to divide by becoming rounded. They then divided longitudinally (fig. 21).

In the same preparation examined thirty-two and a half hours after it was made, agglutination* of the small trypanosomes was seen at a spot near the edge of the drop of blood.[6] The parasites were attached together by their flagella (fig. 22). Their movements had slowed down, and later they were absolutely quiescent. Other

* This phenomenon was seen up to 4 days after the preparation of a blood film.

parasites, rounded and attached to one another as in fig. 23, were seen in this situation. These also became motionless.

Four days later in the same preparation these small parasites were still very active; no change having occurred in them, with the exception of the production of a bulb found at the flagellar end of the herpetomonas-like forms[6] (fig. 24). Five days later many small parasites were rounded and granular, but were still moving. Six days

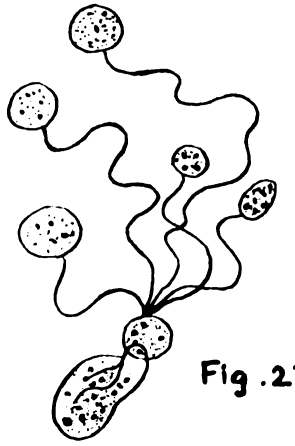


Fig. 23.

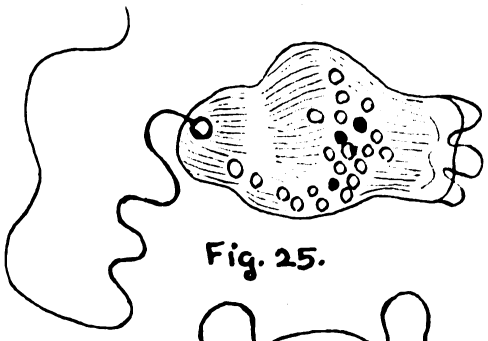


Fig. 25.

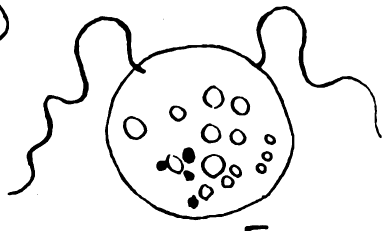


Fig. 26.

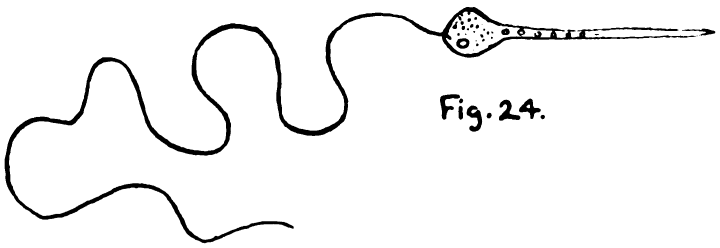


Fig. 24.

later the preparation was discarded, as the small parasites had become rounded, granular and motionless, and the haemoglobin of the red corpuscles was laked.

The next day after the preparation was made a curious looking flagellate was seen. It has large highly-refractile granules, and apparently two flagella; the posterior one moved slowly, the anterior one was fairly active, but the parasite remained stationary (fig. 25). A day later this parasite had become rounded and the flagella were still moving slowly (fig. 26). The following day the parasite had become coarsely granular and the flagella had disappeared.

* Because of their motion and great length it was almost impossible to decide whether there were really two flagella or only one flagellum with a very ample undulating membrane.

Some of the trypanosomes became rounded, but did not develop further than the first or second division. Their outline became indistinct, and, instead of fine granulations, clumps of rather coarse granules appeared, and later showed Brownian movement.

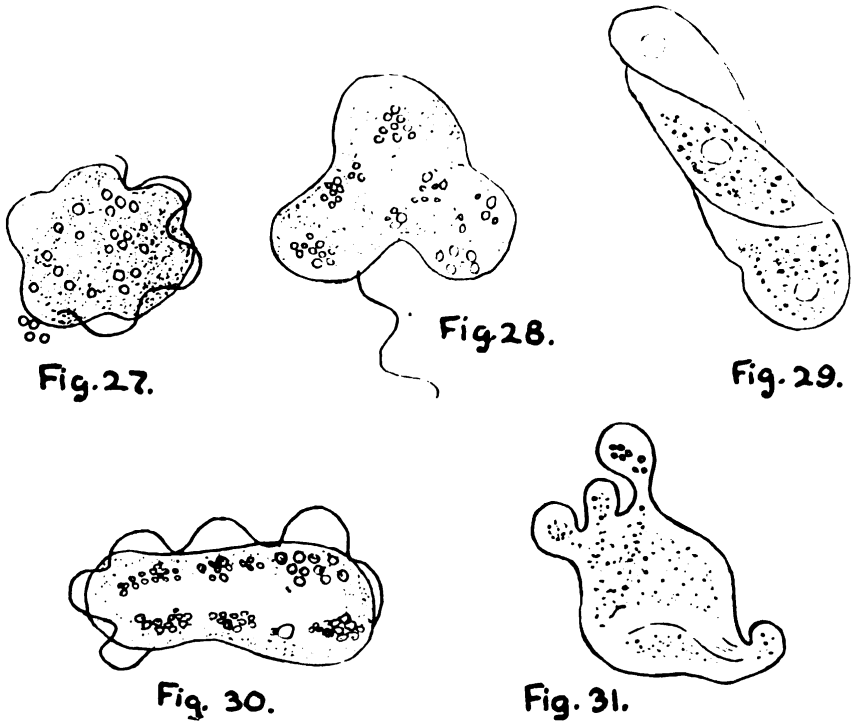
Examples of such parasites watched for five days are as follows:—

Fig. 27.—Shows a coarse granular rounded form.

Fig. 28.—A tri-lobed leaf-like form.

Fig. 29.—A form with fine dancing granules; on the fifth day the granules became large and yellow and the organism disappeared.

Fig. 30.—A coarse granular form without movement. In twenty-four hours there were large refractile granules, apparently fat globules. The flagellum was not so easily seen.



In one preparation an amoeboid form of trypanosome was seen (fig. 31). It contained a few rather coarse granules. On the second day the granules were somewhat coarser, and the parasite had slightly changed its shape, while the pseudopodia had retracted. On the fifth

day it was obviously degenerated. Some of the herpetomonas-like forms were seen to divide longitudinally. The further development of *T. loricatum* was not observed in fresh preparations.

Stained specimens

From the above-named species of frogs and toads, 220 dried films of blood and organ juices stained by our usual Romanowsky method were examined. *T. loricatum* vel *costatum*[2] was found in most of them (Plate XXV, figs. 1, 2, 3 and 4). The measurements of this form are as follows:—

I.—12 to 42 μ.
II.—The nucleus and blepharoplast are apparently connected.
III.—8·2 to 16 μ.
IV.—23·7 to 26 μ.
V.—8·2 to 16 μ.
VI.—22 to 45 μ.
Total length of the parasite, 52 to 101 μ.

It is a curious coincidence that in the twenty parasites measured, the length of the nucleus is approximately the same as that of the flagellum.

In an organism of this size and shape, spreading and drying in the preparation of the film will inevitably produce a certain amount of distortion (Plate XXV, fig. 1). However, the shape of the *T. loricatum* met with in Plate XXV, fig. 3, corresponds with our description of the parasite seen in fresh blood (page 307). The body is ovoid, the posterior extremity being narrower than the anterior. The organism may be found lying at full length, or with one end partly turned over, or completely doubled upon itself. The structure of the cytoplasm is finely granular. The periplast is pleated into ridges which run longitudinally from the posterior to the anterior extremity (Plate XXV, fig. 4). This appearance can occasionally be seen in the most beautiful manner in slightly disorganised slowly moving parasites, in fresh preparations. Occasionally the organism is twisted or folded upon itself, when the ridges consequently seem to run obliquely. The blepharoplast is usually very small and consists of four or more granules imbedded in a matrix. The thickened edge of the undulating membrane takes its origin from it and the nucleus is connected with it (Plate XXV).

The nucleus in nearly every case is elongated and crescentic, with its concave side toward the thickened edge of the undulating membrane*, and its anterior extremity is pointed. It is 1·6μ to 2·7μ wide. In a very few cases it is round, when its diameter is from 3·3μ to 4·4μ (Plate XXV, fig. 3). The undulating membrane is well developed and runs in folds from the blepharoplast across the middle of the body, from the edge of which it extends about 2·7μ before the free flagellum begins. The structure of the nucleus is more or less complex. Its edges are sharply defined, and at its middle are many very small red granules. The anterior portion of it is dense and finely granular and often contains larger masses of chromatin. In some instances large masses of chromatin are found at both ends (Plate XXV, fig. 2). In the fresh this was found to precede division (see figs. 10 and 11 in text).

Numerous forms occur which have a round body, round nucleus, blepharoplast close to the nucleus and a short white line extending from the blepharoplast, like a short flagellum; this line, however, never extends beyond the edges of the parasite. The line resembles the tongue seen in fresh specimens; both occur in rounded parasites of about the same size, but we are unable to state their identity. It is, however, quite possible that this type of parasite may be a developmental form occurring at about this period and that it may be a product of the first dichotomous division of *T. loricatum*. Differentiated areas of obscure nature are sometimes seen in the nucleus, but in none of our specimens have we observed the interesting nuclear changes described by Franca and Athias.[2]

The observations by Moore and Breinl[8] seem to be of interest in connection with the peculiarly elongated "nucleus" of many forms of *T. loricatum*.

In stained smears of kept blood from the same frog, the same cycle of multiplication and development can be followed as mentioned above in freshly made preparations of fluid blood. In Plate XXV, fig. 8, is shown a parasite rounded and about to cast off its flagellum.

* The resemblance of this parasite to those described under other names than *T. loricatum* by Martin (8) in an African lizard, and by Marchoux and Salimbeni (5) in a frog (*Hyla*) is very evident. The parasite described by the latter authors is peculiar in that it had no free flagellum (the flagellum of *T. loricatum* is often stained with difficulty, or may lie out of sight beneath the body of the parasite), and its undulating membrane was commenced by a rigid spur which was observed in *T. loricatum* neither by Franca and Athias (2) nor by ourselves.

The body measures 18μ by 13μ, and the free flagellum was 45μ long The blepharoplast is of the usual structure with the usual clear space about it, and from it extends the nucleus as a band 1·1μ wide and 9μ long.

In Plate XXVI, fig. 14, is shown a round form with the flagellum cast of. The cytoplasm is coarsely granular and the blepharoplast which is of the usual structure is situated near the edge of the body. The nucleus adjoins the blepharoplast on one side. The diameter of the organism is 22μ. The nucleus is about 3·3μ in diameter.

A division, probably the first of a round form, is seen in Plate XXVI, fig. 15. The dividing parasite measures 19μ by 13μ. A group of 16 parasites is shown in Plate XXVI, fig. 18; each of these cells measures about 3·3μ in diameter. Other small parasites, probably representing parasites of a third division (Plate XXVI, figs. 16 and 17), have a diameter from 4·4μ to 6·6μ. Plate XXVI, fig. 15, is a division of forms with two blepharoplasts and two nuclei. Such parasites were frequently seen. Occasionally round cells are found with multiple nuclei and blepharoplasts, but with the cytoplasm undivided (Plate XXVI, fig. 19). Such cells were from 11 to 22μ in diameter.

The small round cell now develops a flagellum from the blepharoplast, and the body elongates in one axis (Plate XXVI, fig. 20). This parasite measures 6·6μ by 5·5μ, and has a free flagellum 22·8μ long. The blepharoplast is 0·8 by 0·3μ, and is 1·1μ from the edge. The nucleus is 1·1μ by 1·6μ.

The next stage in the development is the formation of herpetomonas-like forms (Plate XXVI, fig. 21). These have a body length of from 9μ to 22μ, divided as follows:—Posterior granule (see below) to posterior extremity 0·5μ to 4·4μ, posterior granule to nucleus 4·2 to 10μ, length of nucleus 2 to 4·4μ, nucleus to blepharoplast (when blepharoplast is anterior) 1·2μ, length of blepharoplast 0·5 to 0·8μ, blepharoplast to the anterior extremity 2·2μ to 5·5μ. The flagellum is from 10μ to 25μ. The width of the body is 1·2μ.

The body is narrow, tapering at both ends to a fine point.

There is no undulating membrane (Bouet[6] describes one as being present). There are from 4 to 16 vacuoles lying between the posterior granule and the blepharoplast. The blepharoplast of the usual granular type lies either at one side of the nucleus or slightly anterior to it. From it the flagellum arises and runs through the

middle of the anterior portion of the body, or, turning a little to one side, is closely applied to the margin of the anterior part of the body. In one specimen a bluish line connected the blepharoplast with the nucleus. The nucleus is long and narrow, sometimes extending completely across the body, sometimes occupying half the width. Just posterior to the nucleus is a vacuole, and often when the nucleus does not occupy the whole width of the body there is another beside it. Near the posterior extremity is a red mass, rather more lightly stained than the blepharoplast, which consists of from one to four granules imbedded in a matrix; this we have called the posterior granule. The size of this mass varies from a small point 1μ in diameter. In one case a blue line was seen to extend from this mass anteriorly, but only for a short distance. It did not connect the posterior granule and the nucleus. One of these herpetomonas-like forms has a large anterior end (Plate XXVI, fig. 23), similar to the bulbous form seen in the fresh preparations (fig. 24 in text). Masses of agglutinated parasites of this type were seen (fig. 22). Two anomalous forms were seen which probably belong to this state. One has a body length of 10μ and a width of $4{\cdot}4\mu$. Numerous filamentous flagella seem to arise from the blepharoplast at one end, and the organism seems to be encircled by an undulating membrane. The other is quite herpetomonas-like except for the presence of four flagella (Plate XXVI, fig. 22). It measures as follows:—Posterior extremity to posterior granule $1{\cdot}2\mu$; posterior granule to nucleus $5{\cdot}4\mu$; length of nucleus $3{\cdot}3\mu$; nucleus to anterior extremity $1{\cdot}2\mu$. The anterior part of the body in the region of the nucleus and blepharoplast is wider, being $2{\cdot}2\mu$ wide. The flagella are 15μ, $4{\cdot}4\mu$, $4{\cdot}9\mu$ and $5{\cdot}5\mu$ respectively.

Similar forms were seen in the fresh preparations. They bear some resemblance to the trichomonas described in the intestine of frogs. The blood was, however, taken and kept with every care, and there was certainly no contamination. These forms certainly occur in the blood; whether they represent a stage in the development of *T. loricatum* or not it is difficult to say.

The next stage in the development is a form resembling *T. inopinatum*[3] (Plate XXVI, figs. 25, 26). This stage is found in fresh blood, in contradistinction to the forms just described, which were found in kept blood alone. The parasites of the *T. inopinatum* type may be divided into two groups to facilitate measurements. The

first group has the blepharoplast close to the nucleus. Its measurements are:—

Posterior extremity to nucleus	5·5 to 10·0μ
Measurement III	1·6 to 2·7μ
„ IV	11·0 to 20·0μ
„ V	11·0μ
„ VI	3·3 to 4·4μ
Total length of the parasite	28·5 to 43·0μ
Width of nucleus	1·6 to 2·2μ

The other form of *T. inopinatum* has the blepharoplast posterior to the nucleus, and measures as follows:—

Measurement I	·8 to 12·0μ
„ II	2·0 to 4·4μ
„ III	2·0 to 2·2μ
„ IV	13·2 to 16·0μ
„ V	12·0μ
„ VI	1·6 to 3·3μ
Total length of the parasite	32·5 to 49·0μ
Width of nucleus	1·6μ

The body of both forms is pointed at both ends, or is sometimes somewhat blunt at the posterior end. The blepharoplast* is of the usual structure and is situated in a plane common to the nucleus, or posterior to it. The nucleus is round and sometimes occupies the whole width of the body, and sometimes not. The posterior part of the body tapers gradually, and at the level of the blepharoplast the body is widest. The undulating membrane is never very full in these forms.

Forms resembling *T. sanguinis*[4] (Plate XXVI, fig. 27) are present. They seem to be a further development of *T. inopinatum*, since every gradation exists between these two forms. The undulating membrane is the most striking feature. Its thickened edge arises from the blepharoplast and extends transversely across and beyond the body to border the wide membrane. The blepharoplast is in the same plane as the nucleus. The measurements are as follows:—

* That is, it consists of 2 to 4 or more granules embedded in a matrix and surrounded by a clear area.

Posterior extremity to nucleus	4·4μ
Measurement III	1·1 to 2·2μ
,, IV	12·0 to 19·0μ
,, V	11·0 to 12·0μ
,, VI	1·1 to 2·7μ
Total length of the parasite	30·2 to 39·0μ
Width of undulating membrane	2·7μ

From *T. sanguinis* there seems to be every gradation to the "leaf-like" trypanosomes of Plate XXVI, fig. 28. These forms occur as frequently as does *T. sanguinis*, and are never seen in slides from which *T. sanguinis* is absent. The posterior part of this leaf-like form tapers toward its round and blunt extremity. This portion stains more deeply and seems to consist of folds tightly folded. At about the junction of the posterior and middle thirds of the body is the round nucleus, and in a common plane is the blepharoplast. The folds of the posterior part of the body gradually unfold until about the middle of the body, where they are completely unfolded. The body then tapers to a sharp pointed anterior extremity. The thickened edge of the undulating membrane arises from the blepharoplast, crosses the body obliquely and continues along the edge of the body at a distance of 1μ from it to the pointed extremity where it becomes the free flagellum. The undulating membrane is not well developed, but the thin edge of the body and anterior portion of the body seemed, in the fresh, to act as an undulating membrane. The measurements are:—

Posterior extremity to nucleus	5·5 to 10·0μ
Measurement III	1·1 to 2·7μ
,, IV	13·2 to 26·0μ
,, V	9·0 to 11·0μ
,, VI, at a level of nucleus	6·6 to 9·0μ
Total length of the parasite	30·0 to 48·0μ

Division forms of this stage were seen.

There was another variety of the leaf-form. The posterior part is serrated at the extremity and the serrations seem to be the points of a petal-like arrangement of the posterior end. The general effect is that of a bud about to open (Plate XXVII, fig. 29).

The leaf-like form seems to gradually change to the second variety of *T. loricatum* described by Franca and Athias.[2] The change is brought about by a shortening of the anterior part of the body and a complete unfolding of its posterior portion. Forms representing this transformation are seen in Plate XXVII, figs. 30 and 31. This completes the cycle from *T. loricatum* to *T. loricatum.**

The first part of this cycle, from *T. loricatum* to the herpetomonas-like forms, was actually observed in a single living parasite which was kept under observation during three days. The second part of the cycle, from the herpetomonas-like forms back to *T. loricatum* is largely based upon the examination of stained preparations of fresh and "kept" blood. We are fully aware of the necessity for caution in determining a developmental process from stained specimens. In the present instance the deductions we have made from our examination of stained preparations have been frequently confirmed, and even supplemented by isolated observations on fresh specimens. Our statement is supported by the observation of Bouet[6] who saw every intermediate stage between rounded forms and adult trypanosomes of the *T. rotatorium* type[2] in his cultures.

We describe several types of trypanosomes which are constantly seen. All are simply developmental variations of *T. loricatum.*

I.—Forms resembling the trypanosome found in *Hyla arborea.*

(*a*) A short variety (probably identical with *T. rotatorium*)

(*b*) A long variety.

(*c*) A wide variety.

II.—A lanceolate form resembling somewhat a type described by Laveran and Mesnil.[3]

III.—Forms resembling *T. mega.*[4]

(*a*) *T. mega.*

(*b*) A coarsely reticulated form.

(*c*) A form with large red granules in it.

IV.—Forms resembling *T. karyozeukton.*

V.—An unplaced trypanosome.

* Gaule (2) evidently saw a considerable part of this cycle, since he believed that the trypanosomes were produced from the white blood corpuscles. He stated that he had seen leucocytes each develop an undulating membrane and a flagellum. He also describes trypanosomes which cast off their motile apparatus and so again became leucocytes. The way in which such a mistake could arise is very apparent. A clump of rounded parasites at the fourth or fifth division bears a very close resemblance to a group of white cells.

I.—(*a*) A typical short form resembling that found in *Hyla aborea* (Plate XXVI, fig. 10) is about 25μ long, 9μ wide and has a flagellum 28μ long. The blepharoplast is situated 2·3μ from the posterior extremity, and 1·2μ posterior to the nucleus. The nucleus is 9μ long. The chromatin was collected in masses at either end of the nucleus, or was diffusely distributed in fine granules throughout its extent. The cytoplasm is a rather loose network, and it seems to be continued into the undulating membrane. The side of the body carrying the undulating membrane thus appeared to be folded into a series of foot-like projections produced by the involutions of the thick and substantial membrane.

This general description applies to the other two forms.

(*b*) The long form is characterised by its great length, 40 to 63μ, not including flagellum which is from 19·2 to 26μ long (Plate XXV, figs. 6 and 7), its comparatively narrow body, between greatest projections 5·5 to 7·2μ, and the appearance of folds or pleats running longitudinally; this was especially marked at the posterior end. Sometimes the body is particularly thick and dark-stained. The length of the nucleus is 22 to 24μ, and the width is 1μ.

(*c*) The wide form (Plate XXVI, fig. 9) looks like the narrow varieties, with the folds unfolded however. It measures as follows:—

Measurement	I	22·0μ
,,	II	Blepharoplast is connected with the nucleus.
,,	III	22.0μ
,,	IV	19·0μ
,,	V	14·8μ
Total body length		63·0μ

All the "Hyla forms" have the thickened edge of the undulating membrane running about 1μ from the wavy edge of the body. A round nucleus was never found in any of these forms.

II.—The lanceolate variety (Plate XXV, fig. 5) of *T. loricatum* has a rather wide and round posterior extremity, from which the body gradually tapers toward the pointed anterior extremity. The body has smooth edges on both sides. The undulating membrane, which is well developed, runs first directly backwards and then curves around the end of the body to run in folds along the median line of the body. There is no cytoplasm in the undulating membrane, as seems to be the case in the preceding "Hyla forms" variety.

The nucleus, a long narrow tube tapering at both ends, especially anteriorly, extends from the blepharoplast toward the anterior end. The length of the nucleus was almost the same as that of the flagellum.*

The measurements of this form are as follows:—

Length of body, 38 to 50μ; width, 10 to 16μ; blepharoplast to posterior extremity 2·2 to 4·4μ; width of nucleus 1·2 to 2μ; length of nucleus 19·8 to 29μ; flagellum, 17·6 to 27μ.

A change in the form of a "leaf-like" trypanosome observed in a fresh preparation is illustrated in figs. 32, 33, 34. The parasite was



Fig. 32.

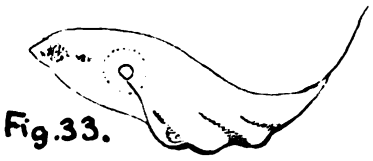


Fig. 33.

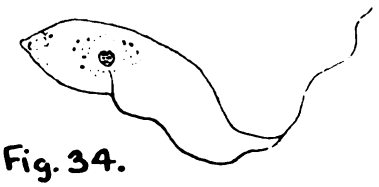


Fig. 34.

watched until it had assumed an appearance, always without well-marked striations, almost identical with *T. mega*. A single form of this type, probably identical with fig. 33, was seen in the stained preparations.

Its measurements are as follows:—

Measurement	I	...	... 9·0μ
,,	II	...	... 2·2μ
,,	III	...	... 4·4μ
,,	IV	...	... 20·0μ
,,	V	...	... 9·0μ
Total length of parasite			... 45·0μ

We look upon this form as an intermediate stage between the "leaf-like" forms (see above) and *T. mega*.[4]

* It is a curious fact that in *T. loricatum* (type) and in two of the varieties described (Ib and II, see page 323) the length of the nucleus was the same as that of the flagellum.

III.—(*a*) The "*Mega*" forms may have long anterior and posterior (Plate XXVII, fig. 39) ends, a contracted posterior extremity (Plate XXVII, figs. 35 and 36), or a short contracted body (Plate XXVII, fig. 37). In fresh preparations the striations are well seen in the long forms;* these may coil up exactly as does *T. karyozeukton* (Plate XXVIII, fig. 43) (see below). *T. mega* has been seen to gradually become more rounded until it became spherical. It then lost its flagellum. The forms with contracted posterior extremity and contracted body (Plate XXVII, figs. 35-37) probably represent stages in this process. From them development was not observed.

In the type of *T. mega* with a short contracted body the length is 40μ, the width at the posterior part 20μ, the nucleus 3·3μ, and the blepharoplast 1·1μ posterior to it. The width of the nucleus varies greatly; in most cases it extends completely across the body, while in others it was at one side and only 2·2μ wide.

T. mega (type) measures as follows—

Measurement	I	..	4·0μ
,,	II	...	1·3 to 6·0μ
,,	III	...	1·1 to 4·4μ
,,	IV	...	15·0 to 53·0μ
,,	V	...	8·2 to 16·0μ
,,	VI	...	3·3 to 11·0μ
Total length of parasite		...	52·0 to 104·0μ
Width of nucleus		...	2·2 to 9·0μ

The original description of *Trypanosoma mega* holds good for these parasites. In a few parasites there was a slight variation in the structure of the nucleus. The clear area just anterior to the nucleus was still present, but the nuclear area of irregular chromatin was replaced by an arrangement of about eight or nine processes which radiated from a focus placed at the middle of the anterior wall of the nucleus. They seemed to be placed in a deep purple stroma, and some of them contained a few reddish granules. A somewhat similar appearance is illustrated by Broden.[7]

(*b*) There was another form similar to the "Mega" type except for the very coarse reticular structure of the cytoplasm (Plate XXVII,

* It may be stated here that more or less distinct longitudinal striations were present in every type of trypanosome mentioned in this paper.

fig. 38). Other forms were quite different. Only five of these well-marked forms were seen. The measurements are as follows:—

Measurement	I	...	... 8·0 to 11·0μ
,,	II	...	... 5·6 to 10·0μ
,,	III	...	... 2·2 to 2·7μ
,,	IV	...	... 15·4 to 21·0μ
,,	V	...	... 13·2μ
,,	VI	...	... 3·0 to 8·0μ
Total length of parasite			... 44·0 to 58·0μ
Width of nucleus		...	... 2·2μ

(*c*) Another form, seen in both fresh and stained preparations, which resembles *T. mega* is a wide trypanosome with large granules scattered through its body. Some had many granules (Plate XXVII, fig. 34), others had few (Plate XXVII, fig. 33). Usually the granules stained a bright red, some of them, however, were refractile and remained unstained. Although the other granules in the same parasite were stained, we can say nothing concerning the origin of these possibly chromidial granules.

These forms measure as follows:—

Measurement	I	...	... 9·0 to 15·0μ
,,	II	...	... 4·4μ
,,	III	...	... 4·4 to 5·0μ
,,	IV	...	... 18·0 to 27·0μ
,,	V	...	... 7·7μ
,,	VI	...	... 6·6 to 10·0μ
Total length of parasite			... 43·5 to 59·0μ

The nucleus in these forms is always pale, and across it can be seen the striations of the body. The blepharoplast is 1·2μ long, rather narrow, and almost hidden by the large granules, which are about 1·5μ in diameter.

IV.—A trypanosome resembling *T. karyozeukton*[4] is present, but the characteristic chain between blepharoplast and nucleus can never be seen. These trypanosomes may be classified, according to the size, as large, medium, and small, or, better, narrow.

The large form (Plate XXVIII, fig. 42) is sometimes coiled more or less tightly (Plate XXVIII, figs. 43 and 44). This phenomenon was also observed in fresh preparations. The further development of these

forms was not seen. The edge carrying the undulating membrane has a wavy outline. The width of the body at the level of the nucleus is less than just anterior or just posterior to it.

The narrow form (Plate XXVIII, fig. 40) was seen in one case to have a blue line running from blepharoplast spirally to the posterior extremity, and the undulating membrane was continued for a short distance posteriorly as a ridge beyond the blepharoplast. The medium form is seen in Plate XXVIII, fig. 41.

All the forms have clear spaces around the nucleus and blepharoplast.

		Large	Medium	Small
Measurement	I	13·2 to 26·0μ	10·0μ	9·0 to 20·0μ
,,	II	7·1 to 13·0μ	6·6μ	3·8 to 8·5μ
,,	III	3·3 to 5·5μ	3·3μ	2·2 to 3·5μ
,,	IV	51·0 to 75·0μ	37·2μ	20·7 to 38·0μ
,,	V	17·6 to 27·0μ	18·7μ	18·5 to 23·0μ
,,	VI	3·3 to 7·7μ	3·8μ	1·6 to 2·7μ
Total length of parasite		102·6 to 134·0μ	76·7μ	59·5 to 87·0μ
Width of nucleus		3·2 to 5·7μ	2·2μ	1·6 to 2·7μ

V.—The remaining trypanosome to be described is a long narrow form, with a narrow nucleus, and a large blepharoplast (Plate XXVI, fig. 24). It measures as follows:—

Measurement	I	 3·3 to 6·6μ
,,	II	 13·2 to 19·2μ
,,	III	 2·2 to 3·3μ
,,	IV	 6·6μ
,,	V	 11·0 to 16·0μ
,,	VI	 1·1μ
Total length of parasite		... 37·0 to 54·0μ

The cytoplasm is rather coarsely reticular, the body is pointed at both extremities, the large blepharoplast is situated in the centre of a clear space, from which arises the rather scanty undulating membrane.

This parasite resembles *T. inopinatum* in many ways, and may be a parasite of that form.

The question arises whether the cycle of multiplication just described is completed in the frog, or whether it normally occurs only outside the frog, probably in a second blood-sucking host, as a leech. The smallest rounded parasites with flagella and the herpetomonas-like forms never occurred in freshly-drawn blood. Bouet[6] agrees with this observation in stating that none of the young parasites were seen in the blood. Franca and Athias,[6] however, record that they saw small rounded parasites both with and without flagella (probably forms of *T. loricatum*) in blood fixed immediately after withdrawal. A single small rounded parasite (6μ in diameter) was seen in our series of slides of freshly-drawn blood. With the exception noted above, every other type of parasite occurring in the developmental cycle described in this paper was observed in fresh-drawn blood. From these observations it seems that this cycle of "swarm" division may be completed in the amphibian host, but that the smallest forms are rarely present in the peripheral blood. It is noteworthy in this connection that there is no certain correspondence between the length of time blood containing *T. loricatum* has been kept and the type of trypanosomes present in it.

More than once large specimens of *T. loricatum* were seen in whose substance red blood cells occurred. Marchoux and Salimbeni believe that the cells and the parasite are merely superimposed. Without wishing to assert that the cell has been ingested, that is, that its presence in the parasite is due to more than a mechanical accident, we are confident that we have seen instances where the cell was definitely within the cytoplasm of the trypanosome. The trypanosomes may be attacked at any stage of their development by leucocytes. Frequently they seem to resist successfully and are not ingested. In one instance a leucocyte was seen to ingest the haemoglobin containing stroma of a disintegrated red cell; it was interesting to note that its protoplasm contained dark brown granules —a most unusual occurrence.

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*Drepanidia.**

Fresh Preparations.

Drepanidia were present in every species of frog and toad named on page 307, and in almost every frog examined. In the same frog (*Rana galamensis*) in which the developmental changes in *T. loricatum* were observed, drepanidia were present in almost every other red blood corpuscle, though scarcely any young parasites were seen. The red cells were swollen to nearly twice their normal size, the colouring matter had disappeared, and the cells in fresh preparations presented the appearance of a crumpled colourless bag, to one side of which the parasite lay. The nucleus was pushed to the periphery of the cell (Plate XXIX, fig. 86).

A few of these drepanidia were found free in fresh preparations examined immediately after making them, but the number of free parasites increased enormously after half an hour. The drepanidia, after leaving the cells and becoming free, were long and had a round anterior end, *i.e.*, the end in advance during progression (Plate XXVIII, fig. 67). A little way from this end the body tapered gradually to the posterior end, which though narrow, was bluntly rounded off (Plate XXVIII, figs. 68 and 69).

Progression takes place in three ways:—

(1) With the body stretched out by a gliding movement.

(2) By gregarine-like contractions of the protoplasm. This occurs when obstruction is encountered (Plate XXVIII, figs. 70 and 71).

(3) By a sudden jerking backward of the posterior end which has previously curved round to meet the anterior end.

* See footnote to page 303.

After about an hour, agglutination of the free parasites was observed. They joined themselves together by their posterior, sharper, somewhat granular ends. Two, four, or six parasites came together in clumps in this way. Contractions occurred from side to side, causing the parasites to bend. Two days after the fresh preparations were made, the agglutinated groups of drepanidia were observed still, the individuals being rather granular and stumpy and actively motile, but they did not undergo any further change.

Three forms of drepanidia were observed, a small form (Plate XXVIII, fig. 74) which will be described in stained preparations, a medium form (Plates XXVIII and XXIX, figs. 82, 68, 69) (the common form), and a very large form (Plate XXIX, fig. 86). These large drepanidia often contained large yellowish, highly refractile granules, which varied in size and number in different parasites. There were from one to about one hundred of these globules in each parasite (Plate XXIX, figs. 84, 85, 89, 90, 91, 92). These highly refractile granules were situated for the most part around the nucleus. In some of these parasites small dancing granules were seen at the more slender end.

Stained Specimens.

The following description is based on the examination of dried and stained films.

The nucleus of the host-cell was displaced (Plate XXVIII, fig. 59) but not injured; the cytoplasm, however, suffers severely since the parasite seems literally to tunnel about within the limits of the cell. Many corpuscles were found in which the contents seemed to have been devoured, leaving them looking like wrinkled empty sacks (Plate XXVIII, fig. 67). Such sacks could be seen with the drepanidia present, or leaving, or gone. Two and three parasites were sometimes seen in one blood corpuscle. The drepanidia were not only present in erythrocytes but were also occasionally found in leucocytes.

As already noted there were three principal forms of drepanidia; a large form, a medium form, and a slender form. The medium form was the most common (Plate XXIX, figs. 81, 82), and was from 15 to 18μ long and from 5 to 6μ wide. It had a fine granular striated protoplasm, which sometimes contained coarse red granules, and sometimes not. Some specimens had a few (four) granules at the anterior end of the body, and some had a straight blue line running

from the nucleus to the anterior extremity. The nucleus was from 3·3 to 4·4μ in diameter, and was placed about 7μ from the anterior extremity and 4μ from the posterior extremity.

One and two constrictions in these forms were observed, showing the method of progression described in fresh specimens (Plate XXVIII, figs. 70, 71). In a few specimens the nucleus had apparently divided into two, and the halves were attached by a red line (Plate XXVIII, figs. 68, 69).

Some of these forms were encysted, both in erythrocytes and free in the serum. Such encysted forms often had a small amount of red-staining excretion at both ends (Plate XXVIII, figs. 59, 60).

The "slender" forms (Plate XXVIII, figs. 73, 74) were from 13 to 15μ long and 1·1μ wide. The cytoplasm was light-staining and of a loose texture. The shorter or posterior end was very faint-staining and the extremity could be seen with difficulty. The longer or anterior end was a faint pink, which deepened towards the extremity. On both sides of the nucleus were areas staining the same as the cytoplasm of the erythrocytes. There were two, sometimes three, of these areas which were from 0·6 to 1·7μ long. Near the posterior extremity there were sometimes from two to four or more red chromatin granules, and occasionally there were a few in the anterior part of the body. The nucleus consisted of eight peripherally arranged chromatic granules, "chromosomes,"* and one or two darker, central chromatic granules, "karyosomes," all connected by fine lines. In one instance (Plate XXVIII, fig. 73) a blue spiral line was found to run from one of the chromatic granules to the anterior extremity of the body. The nucleus was 2·2μ long and was placed 9μ from the anterior extremity, and 3·3μ from the posterior extremity. This "slender" form may be present in the blood with the larger form, or it may be present in blood which has none of the larger forms.

These "slender" drepanidia penetrate the erythrocytes, then lose both ends, so that only the nucleus and a small amount of cytoplasm around it is left. The chromatic granules go to the side, and later they are gathered in a mass at one end of the body, with a few chromatic granules remaining outside the mass (Plate XXIX, fig. 76). The body of the parasite enlarges and at the extremity opposite the chromatin mass are several round clear spaces, which may or may

* It must be understood that the terms chromosomes and karyosomes are used in a purely descriptive sense.

not contain a granule of chromatin. The mass of chromatin divides into two, and the cytoplasm may or may not divide synchronously. In cases where the cytoplasm does not divide synchronously, the chromatin goes on dividing until there are from 10 to 16 masses of chromatin (Plate XXIX, figs. 78-80) arranged almost around the edge of the cell. These rosettes later show divisions into young parasites (Plate XXIX, fig. 79). Rosettes were seen leaving the host-cell, and also found free in the serum (Plate XXVIII, fig. 66). In one slide a group of eleven young drepanidia that had just escaped from the rosette condition was seen (Plate XXVIII, fig. 72). These forms were 7·7μ long and 1·1μ wide. Their cytoplasm was of a faint-staining, coarse, granular structure. The nucleus was composed of a larger, central mass of granules and, usually, a chromatin granule on either side; occasionally both were on one side. In one case the chromatin seemed to be dividing (Plate XXVIII, fig. 72). These sporulating* forms always arose from the "slender" drepanidia. In one series of eight slides from one frog many sporulating and "slender" forms were seen, but none of the larger drepanidia described in the next paragraph.

In a few frogs, *Rana mascariensis*, a very large drepanidium was seen, which was like the common form, but larger. It was lighter stained and often took a pink colour instead of the normal blue. The cytoplasm was coarsely granular, and many of the parasites had non-staining, highly-refractile globules in them. These globules were from one to one hundred in number, and from 0·6μ to 7·7μ in diameter. They were always found near the nucleus and gave the appearance of a fatty degeneration of the nucleus. They sometimes distended the drepanidium until it was nearly round (Plate XXIX, fig. 89), but they usually did not affect the size of the parasite. The large type of drepanidia presented three forms, a long form, a short or young form, and a folded or "two-shanked" form. The long form (Plate XXIX, fig. 86) was usually 22·6μ long and 7μ wide. The nucleus was 3·4μ long and 5·6μ from the posterior extremity and 13·6μ from the anterior extremity.

The short, or young form (Plate XXIX, fig. 83) was about 8·8μ long and 6·6μ wide. The nucleus was 4·4μ in diameter and placed in the centre of the organism.

The folded form was seen in the various stages of unfolding (Plate XXIX, figs. 87, 88).

* The term is not used in its specific sense.

Cytamoeba.

Cytamoebae were present in a few of the frogs in considerable numbers. Amongst the forms seen was the round form (Plate XXIX, figs. 104, 106, 108, 110) previously described in the snake (page 304) (2·2μ wide). In it were short rods, or sometimes round, red dots (Plate XXIX, fig. 104). Other forms were oval or finely granular, sometimes with no apparent internal structure (Plate XXIX, fig. 103). The round forms were often found near the nucleus, and sometimes apparently emerging from it, giving the appearance of fragmentation of the nucleus (Plate XXIX, fig. 110). Some of the parasites were filled with slender rods, which at times projected beyond their edge for a distance of 4μ (Plate XXIX, figs. 112, 113). Other forms were present which looked like masses of rods bound together (Plate XXIX, figs. 107, 108, 109, 115); sometimes these bundles of rods were free in the serum (Plate XXIX, fig. 100). Occasionally short rods were found free in the erythrocytes, sometimes end to end, sometimes crossing one another at right angles. The rods found in all the above forms were of two kinds, a short rod with rounded ends, and a long filamentous rod. Some of the short rods were seen free in the blood plasma in such forms and arrangements that there seems to be no doubt that some of them at least are bacteria. It is equally certain that some of the longer slender rods are merely filamentous cytoplasmic processes of the rounded parasite. In some of the specimens the body of the cell was stained a light blue, while the rods within were stained a bright red.

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An unidentified parasite

Another parasite, a round, red-staining mass from 1·5 to 5·5μ in diameter, is found in red cells (Plate XXIX, figs. 93, 95, 99). With it are associated bluish-green, crystal-like bodies (Plate XXIX, figs. 93, 95, 97, 98). Both the red masses and crystals may be present in varying numbers (Plate XXIX, figs. 98, 99). The cytoplasm of the host-cells is usually stained a deeper blue than usual. Many of the smaller red masses have a well-defined blue area around them, which suggests that this parasite possesses a cytoplasmic body (Plate XXIX, figs. 94, 96). In stained films of fresh blood the red masses do not seem to have much structure, but in films of blood kept for two days, the structure consists of chromatic granules arranged peripherally with one or two darker chromatic granules in the centre.

This parasite with its crystals was seen in fresh specimens. It occurred in conjunction with drepanidia and *T. loricatum.*

FISHES*

Fresh-water fish of many different sorts were examined in the Gambia and in the Congo. Parasites were found only in the fish without scales, mentioned below.

Those infected were caught in a very small sluggish stream with a very dirty bottom; none of them were over seven inches in length and none were infected with drepanidia.

Trypanosomes (Plate XXX).

Two "mudfish" (*Clarias angolensis*) caught at Leopoldville in the Congo Free State on December 9th and December 30th, 1903, had actively motile trypanosomes in their blood which may be divided into three types; a small, a medium, and a long form. The small type was identical in both, the medium was present in both, but with slight differences, and the long form was present only in the fish of December 9th. The small form was very frequently seen in the fish of December 30th, while in the fish of December 9th, only four were present in a total of ninety-nine parasites. The medium form, in the fish of December 30th, was less frequently met with than the small form, but was often seen in the fish of December 9th. Of the long forms only thirteen were seen. All the forms were characterised by the possession of a large four-lobed blepharoplast,

* The description of the parasites found in fishes is republished by permission from the Journal of Medical Research.

situated at, or very near, the posterior extremity, and by a clear space about the nucleus. The blepharoplast apparently consisted of four darkly-staining granules of equal size embedded in a matrix. In a few specimens there were, perhaps, more than four granules in the blepharoplast, but in these cases it was never possible to be certain of the exact number. The three forms of parasites seemed but different variations of one species, as gradations between all forms could be seen.

The small type was readily distinguished by its size, its narrow body, its relatively long nucleus situated more anteriorly than in the other two types, and with its long diameter parallel to that of the body and by its faint-staining reaction. The blepharoplast was oval, or round, large and distinct (measuring 1·13 by 0·75μ), and was situated at the posterior extremity. The undulating membrane was well developed, having a width of 0·75μ. The long, oval nucleus was situated at the junction of the middle and anterior thirds of the body. It was granular in structure and contained a fibrous network on which were seen eight to sixteen chromatin granules, and one darker granule—the karyosome. The body protoplasm was reticular, and contained a few violet-staining granules. Occasionally parasites were seen in which the body was filled with these coarse chromatophilic granules. Superficial longitudinal striations to the number of five could be seen near the nucleus of two slightly injured parasites. Division forms were very numerous in this type (even when but four were seen in the fish of December 9th, one of them was dividing). They followed the usual method of longitudinal division. In the division of the blepharoplast, the posterior lobe on the concave side of the parasite moved behind the other posterior lobe; the axis of the posterior lobes thus became longitudinal, while that of the anterior lobes remained transverse. The two posterior lobes then separated from the anterior lobes and their axis became transverse again.

The medium type was characterised by its size, and its darker staining reaction. The blepharoplast was large, oval or round, distinct and measured about 1·5 by 0·75μ. It was situated at the posterior extremity, or from 1 to 1·5μ anterior to it. The flagellum was relatively longer than in the short form. The undulating membrane in parasites from the fish of December 30th was well

developed, and wound in numerous folds around the body; it was 1·5 μ wide. In trypanosomes from the fish of December 9th, the undulating membrane was poorly developed, had a few folds and was not more than 1 μ in width. The nucleus was round to oval, its long diameter was at right angles to that of the body, and it occupied the whole width of the parasites. It was granular in structure and contained a fibrous network on which were seen sixteen to twenty-four chromatin granules and one or two darker granules—the karyosomes. The body was more darkly stained in the fish of December 30th. Superficial longitudinal striations to the number of seven were counted near the nucleus. Divisional forms were few in this type and followed the usual method of longitudinal division.

The long type was characterised by its size, its tapering extremities, its comparatively short flagellum and by the possession of from one to four vacuoles just anterior to the blepharoplast. The blepharoplast was large, oval or round, and situated 2 μ from the posterior extremity; its measurements were the same as those of the medium type. The nucleus had the same characteristics as that of the middle type, except that it did not quite occupy the width of the body. The protoplasm was the same as in the medium type, except for the above mentioned vacuoles, which were always present. Eight superficial longitudinal striations were present and these were seen to cross the nucleus. No divisional forms of this type were seen.

The measurements of the three types were as follows:—

	Small form	Medium form	Long form
Measurement I	... 0·0 μ	... 0·0 to 1·5 μ	... 2·0 μ
,, II	... 12·7 μ	... 14·2 μ	... 25·0 μ
,, III	... 3·0 μ	... 3·7 μ	... 45·0 μ
,, IV	... 6·7 μ	... 12·0 μ	... 19·5 μ
,, V	... 12·0 μ	... 13·5 μ	... 10·5 μ
,, VI	... 1·0 μ	... 3·0 μ	... 4·5 μ
Total length ...	... 34·5 μ	... 45·0 μ	... 61·5 μ

Although the trypanosome described by Montel[1] closely resembles our large form, we have seen no account in the literature of any parasite morphologically identical with those described above.

LITERATURE

Good lists of the publications on the trypanosomata of fishes will be found in the works of Laveran and Mesnil, "Trypanosomes et Trypanosomiasis," and of Mense, "Handbuch der Tropenkrankheiten."

The following are important papers not mentioned in these volumes:—

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2. LEBAILLY. Recherches sur les Hématozoaires des Téléostéens marins. Thèse Fac. Med. Paris, 1906, p. 76; and Archiv. de Parasitologie, T. X, 1906.
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5. BRUMPT. Mode de transmission et évolution des Trypanosomes parasites des poissons. Comptes Rendus de la Société de Biologie, 17th Jan., 1906, T. LX. No. 4, p. 162.
6. BALFOUR. Second Report of the Wellcome Research Laboratories, Khartoum, page 197.

A Spirochæte (Plate XXXI).

In the blood of the fish of December 30th a spirochaete-like organism was found which could be identified with no previously described spirochaete.

It had a hair-like body; its length was usually 18μ; its breadth at the widest part was 0·6μ. It was widest at one extremity or towards the middle; in the latter case, the parasite tapered towards both ends, but one extremity was always much thicker than the other. The wider end was rounded, while the other end was drawn out to a fine and slender point. The parasite usually occurred singly and lay in a simple curve, forming three-quarters of the circumference of a circle (Plate XXXI, figs. 1 and 2). Other forms—coils (fig. 3), compound curves (figs. 4, 9-12) and spirals (figs. 5-8) were seen as illustrated. The coils were always formed by the slender end of the parasite. The spiral forms were long, measuring from 24-27μ, and in some of these forms there was a suggestion of a membrane (figs. 5 and 6). The parasites were also seen in pairs and in groups of from three to many individuals as shown in figures 13, 14-16. These organisms seemed to consist of a deeply-staining core and a lightly-staining periplast. Many of them stained irregularly by modifications of Romanowsky's method. The lighter stained areas (fig. 2) occurred at irregular intervals and were of

irregular extent. The periplast could be seen at these lightly stained areas and it was continued to form the slender extremity from which the core was also absent. Occasionally granular forms were seen in which the protoplasm of the core apparently arranged itself into small granular masses, but no regularity could be detected in the arrangement of these granules. Undoubted multiplication forms were not seen.

For descriptive purposes we propose for this organism the name of *Spirochæta jonesii.*

ARTHROPODA

The protozoa found in this class were encountered by chance during an investigation of the development of various haematozoa, while examining various blood-suckers, either as controls, or for the presence of developmental forms of the blood parasites. Many tsetse flies and mosquitoes, a few *Stomoxys*, ticks of various sorts, fleas and lice, and "Congo floor maggots," were examined. The only protozoan parasites observed are recorded below.

MOSQUITOES

(1) *Myzorhynchus paludis.*

A cluster of radiating club-shaped bodies (Colonie radiée) was seen in the thoracic tissues of a female of the above species, examined for malaria at Lusambo. The parasites became free, developed a flagellum and were actively motile in the salt solution in which the mosquito was dissected.

(2) *Pyretophorus costalis.*

Some excitement was at first created during the examination of the alimentary canal of mosquitoes, fed at varying antecedent periods on animals infected with trypanosomes, by the presence of vermicule-like bodies (Plate XXXII, fig. 14), measuring about 18 by 2·2μ. These parasites progressed slowly by active lashing and slower amoeboid movement; some similar forms were motionless.

The amoeboid movements were of two sorts. The first involved a change in the whole shape of the parasite. The second was by a simple protrusion of protoplasm from the rounded anterior, never from the effilated posterior extremity.

They occurred either within the stomach, in the stomachal tissues, or in the body cavity of the mosquito. A central differentiated area could be detected in them by examination in fresh specimens; near the area were more or less numerous granules which often exhibited lively Brownian movement. After some time the parasites became much stumpier (8μ by 16μ), and were later almost spherical. One parasite, however, remained unchanged and active for twenty-four hours.

Further search showed that the parasite occurred in freshly-caught and freshly-hatched mosquitoes, and in the larvae from which all our experimental mosquitoes were raised, and it was quickly shown to be a stage in the life cycle of a coccidium. It was most interesting that practically every larva from one pool was infected with this coccidium, while none of the larvae and adults (anopheline and culicine) from a pool only a few yards distant were affected. So far as was observed the parasite did not occasion any excessive mortality amongst the mosquitoes infected. This seems difficult to understand—the mosquitoes were probably not watched long enough—since the tissues of many of them were fairly riddled with coccidium cysts. The water of the heavily-infected pool was centrifugalised and examined. Coccidium cysts were not recognised, various infusoria and a clump of herpetomonas-like flagellates were alone seen.

So soon as it was certain that the vermicule-like "sporozoite" had no connection with the trypanosomes ingested, the study of this coccidium was discontinued. It is, therefore, not possible to say whether the forms observed represent stages in more than one parasite or no. It seems probable that but one species of coccidium was present. We describe the forms observed and indicate the position they seem to occupy in the life cycle of the parasite. The mobile sporozoites first seen become free through the rupture of a sporoblast containing eight sporozoites. The number of sporozoites in each sporoblast is almost invariable. Fig. 15 is unusual in that the nucleus of two of the sporozoites has divided, and in one instance division of the cytoplasm seems to have commenced. As a rule the long diameters of the sporozoites are parallel. These are, therefore, regularly arranged like the segments of an orange (corps en barillet). Single sporoblasts surrounded by a definite cyst wall may occur. They are more usually seen in groups of four, seven, or even as many

as eleven sporoblasts packed together in an oocyst with a definite capsule. The individual sporozoites have no capsule. Their cytoplasm stains a light blue by Romanowsky's method. It is alveolar in structure and rarely contains granules of any sort, but may have occasional granules. The nucleus is placed centrally, is loose in texture and consists of a varying number of chromatin granules placed in a more lightly-staining matrix. From the analogy of other coccidia, we assume that the forms just described represent the completion of sporogony. The male and female gametes, which probably commence this cycle, and their conjugation have not been seen. Neither has the further development of the sporozoites been observed.

The number of rounded merozoites (figs. 16, 17, 18), present in every part of the larvae and adults dissected, was often extraordinary. These organisms measured about 4 to 5μ in diameter. They were either free or enclosed in a host cell. On becoming extracellular they were seen to possess a definite capsule (pink-staining); this was quickly discarded (figs. 18, 19) and the empty shells were very frequently seen. Often merozoites which had just cast their capsules occurred in groups of four (fig. 18). Four merozoites were, however, never seen within an unbroken capsule. As a rule whether intra or extra-corpuscular the merozoites occurred singly. When extra-corpuscular they were occasionally in pairs. Sometimes large intra-corpuscular cysts were seen which contained very large numbers of merozoites. The merozoites (figs. 16, 17, 18) consisted of a pinkish alveolar cytoplasm in which was situated a nucleus usually surrounded by an indefinite bluish area. In this bluish area a lighter, rounded spot was almost invariably present. A few chromatophilic granules and small clear spaces were frequently seen in the cytoplasm. The nucleus consisted of irregular masses of chromatin placed in a matrix. In some forms (fig. 16) the granules were placed at the periphery of the nucleus and were joined by threads with a central darker-staining mass. In fresh specimens they were immobile.

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TSETSE FLIES (*Glossina palpalis*)

Parasites of the type described below were found in specimens of *Glossina palpalis* freshly-caught, and at all periods, up to 11 days, after having fed on known animals infected with trypanosomes. All the parasites were not precisely similar, but one description will suffice for all. The free rod-like parasites were found only in the alimentary canal, nearly all of them in its anterior part. They were most numerous near the oesophagus, in the "stomach" or in the neighbouring part of the intestine. Cysts containing them were seen attached to, or actually in, the wall of the intestine. Some of them were whole, others had burst (or were burst by slight pressure), and from them were expelled the rod-like bodies. In fresh preparations these rods (Plate XXXII, figs. 1, 2, 3, 4, 5, 8, 9, 10) were non-motile, they contained one or two refractile granules and they occurred singly or, occasionally, in small clumps. The most usual form seen in stained specimens was a rod about 7·0μ in length and 1·0μ in width with abruptly rounded ends (figs. 1, 2, 5). It was limited by a definite outline, but apparently not by a capsule. Its body substance consisted of pale blue protoplasm in which occurred chromatophilic granules and rounded clear areas. The chromatophilic granules varied in size from extremely fine particles (fig. 6) up to masses measuring about 0·3 to 0·5μ in width (fig. 3). They were usually distributed more or less regularly in transverse bands (figs. 1, 2, 5, 10) lying, at intervals, across the parasite. In such parasites the individual granules could barely be distinguished. Often between each band of granules was placed one of the rounded, clear areas (figs. 1, 2, 5, 10). Such parasites were probably preparing for transverse division, as small forms, apparently consisting of one or two rounded segments of the original rod-like body, were often observed (fig. 7). In some of the parasites showing the band-like distribution of chromatin, as well as in others in which pairs of granules were arranged in bilateral symmetry (figs. 8, 9), it seemed as though longitudinal division might be about to occur. In many parasites the chromatin granules were distributed absolutely without order (figs. 3, 4, 6). Rarely a larger and a smaller granule seemed to be in connection with each other.

Rod-shaped parasites of all these three types were frequently terminated by a small chromatophilic granule (figs. 4, 5, 10) (sometimes placed in a clear area), or by a rounded enlargement which

often measured twice the diameter of the rest of the parasite (fig. 10). Rounded parasites measuring from 3μ to 5μ or more in diameter with irregularly distributed granules and no clear areas occurred (figs. 11 and 13). The smaller rounded forms can divide transversely; apparently after a concentration of the chromatin (fig. 12).

It seemed possible that this parasite should be placed amongst the *Myxosporidia*. In recognition of the collaboration of Dr. Inge Heiberg in the work of our expedition, and in particular of his observations on *T. loricatum*, we suggest for the name of this parasite *Myxosporidium heibergi*. About half of the tsetses dissected at Leopoldville had these parasites free in their midgut. In the stomach wall of one freshly caught fly coccidian-like cysts were seen. (" Corps en barillet " ; compare the parasite of *Pyretophorus costalis* described above).*

EPILOGUE

Although the facts presented in this paper are admittedly the results of passing observations, incomplete, disconnected, and made under unfavourable circumstances, we think that they possess a certain value as a record alone of protozoan infections encountered by chance during three years' work in Africa. Many of the appearances noted, though incomplete in themselves, are of additional value in that they confirm the constancy of morphological changes noted in other allied and better studied parasites.

The following examples are cited. Longitudinal striations have been seen in every trypanosome examined; the myonemes of other authors are, therefore, constant in the trypanosomes we have dealt with. In several instances the thickened edge of the undulating membrane of trypanosomes has been observed to terminate, not in the blepharoplast, but in a lighter-staining pinkish granule, or pair of granules, closely applied to it. In some of our trypanosomes the structure of the nucleus and the occurrence, in neighbouring differentiated areas, of paired chromatophilic granules suggests a connection with the complicated nuclear developmental phenomena observed in trypanosomes by some observers. The clear, unstained

* This is the last of the parasitic protozoa observed during our expedition to the Gambia and the Congo. The filariae found in various mammals, birds, amphibians and reptiles will be considered in a forthcoming paper by Professor. H. E. Annett.

III.—(*a*) The "*Mega*" forms may have long anterior and posterior (Plate XXVII, fig. 39) ends, a contracted posterior extremity (Plate XXVII, figs. 35 and 36), or a short, contracted body (Plate XXVII, fig. 37). In fresh preparations the striations are well seen in the long forms;* these may coil up exactly as does *T. karyozeukton* (Plate XXVIII, fig. 43) (see below). *T. mega* has been seen to gradually become more rounded until it became spherical. It then lost its flagellum. The forms with contracted posterior extremity and contracted body (Plate XXVII, figs. 35-37) probably represent stages in this process. From them development was not observed.

In the type of *T. mega* with a short contracted body the length is 40μ, the width at the posterior part 20μ, the nucleus 3·3μ, and the blepharoplast 1·1μ posterior to it. The width of the nucleus varies greatly; in most cases it extends completely across the body, while in others it was at one side and only 2·2μ wide.

T. mega (type) measures as follows—

Measurement	I	..	4·0μ
,,	II	...	1·3 to 6·0μ
,,	III	...	1·1 to 4·4μ
,,	IV	...	15·0 to 53·0μ
,,	V	...	8·2 to 16·0μ
,,	VI	...	3·3 to 11·0μ
Total length of parasite		...	52·0 to 104·0μ
Width of nucleus	...	...	2·2 to 9·0μ

The original description of *Trypanosoma mega* holds good for these parasites. In a few parasites there was a slight variation in the structure of the nucleus. The clear area just anterior to the nucleus was still present, but the nuclear area of irregular chromatin was replaced by an arrangement of about eight or nine processes which radiated from a focus placed at the middle of the anterior wall of the nucleus. They seemed to be placed in a deep purple stroma, and some of them contained a few reddish granules. A somewhat similar appearance is illustrated by Broden.[7]

(*b*) There was another form similar to the "Mega" type except for the very coarse reticular structure of the cytoplasm (Plate XXVII,

* It may be stated here that more or less distinct longitudinal striations were present in every type of trypanosome mentioned in this paper.

fig. 38). Other forms were quite different. Only five of these well-marked forms were seen. The measurements are as follows:—

Measurement	I	...	8·0 to 11·0μ
,,	II	...	5·6 to 10·0μ
,,	III	...	2·2 to 2·7μ
,,	IV	...	15·4 to 21·0μ
,,	V	...	13·2μ
,,	VI	...	3·0 to 8·0μ
Total length of parasite		...	44·0 to 58·0μ
Width of nucleus		...	2·2μ

(*c*) Another form, seen in both fresh and stained preparations, which resembles *T. mega* is a wide trypanosome with large granules scattered through its body. Some had many granules (Plate XXVII, fig. 34), others had few (Plate XXVII, fig. 33). Usually the granules stained a bright red, some of them, however, were refractile and remained unstained. Although the other granules in the same parasite were stained, we can say nothing concerning the origin of these possibly chromidial granules.

These forms measure as follows:—

Measurement	I	...	9·0 to 15·0μ
,,	II	...	4·4μ
,,	III	...	4·4 to 5·0μ
,,	IV	...	18·0 to 27·0μ
,,	V	...	7·7μ
,,	VI	...	6·6 to 10·0μ
Total length of parasite		...	43·5 to 59·0μ

The nucleus in these forms is always pale, and across it can be seen the striations of the body. The blepharoplast is 1·2μ long, rather narrow, and almost hidden by the large granules, which are about 1·5μ in diameter.

IV.—A trypanosome resembling *T. karyozeukton*[4] is present, but the characteristic chain between blepharoplast and nucleus can never be seen. These trypanosomes may be classified, according to the size, as large, medium, and small, or, better, narrow.

The large form (Plate XXVIII, fig. 42) is sometimes coiled more or less tightly (Plate XXVIII, figs. 43 and 44). This phenomenon was also observed in fresh preparations. The further development of these

forms was not seen. The edge carrying the undulating membrane has a wavy outline. The width of the body at the level of the nucleus is less than just anterior or just posterior to it.

The narrow form (Plate XXVIII, fig. 40) was seen in one case to have a blue line running from blepharoplast spirally to the posterior extremity, and the undulating membrane was continued for a short distance posteriorly as a ridge beyond the blepharoplast. The medium form is seen in Plate XXVIII, fig. 41.

All the forms have clear spaces around the nucleus and blepharoplast.

		Large	Medium	Small
Measurement	I	13·2 to 26·0μ	10·0μ	9·0 to 20·0μ
,,	II	7·1 to 13·0μ	6·6μ	3·8 to 8·5μ
,,	III	3·3 to 5·5μ	3·3μ	2·2 to 3·5μ
,,	IV	51·0 to 75·0μ	37·2μ	20·7 to 38·0μ
,,	V	17·6 to 27·0μ	18·7μ	18·5 to 23·0μ
,,	VI	3·3 to 7·7μ	3·8μ	1·6 to 2·7μ
Total length of parasite		102·6 to 134·0μ	76·7μ	59·5 to 87·0μ
Width of nucleus		3·2 to 5·7μ	2·2μ	1·6 to 2·7μ

V.—The remaining trypanosome to be described is a long narrow form, with a narrow nucleus, and a large blepharoplast (Plate XXVI, fig. 24). It measures as follows:—

Measurement	I	 3·3 to 6·6μ
,,	II	 13·2 to 19·2μ
,,	III	 2·2 to 3·3μ
,,	IV	 6·6μ
,,	V	 11·0 to 16·0μ
,,	VI	 1·1μ
Total length of parasite		... 37·0 to 54·0μ

The cytoplasm is rather coarsely reticular, the body is pointed at both extremities, the large blepharoplast is situated in the centre of a clear space, from which arises the rather scanty undulating membrane.

This parasite resembles *T. inopinatum* in many ways, and may be a parasite of that form.

The question arises whether the cycle of multiplication just described is completed in the frog, or whether it normally occurs only outside the frog, probably in a second blood-sucking host, as a leech. The smallest rounded parasites with flagella and the herpetomonas-like forms never occurred in freshly-drawn blood. Bouet[6] agrees with this observation in stating that none of the young parasites were seen in the blood. Franca and Athias,[6] however, record that they saw small rounded parasites both with and without flagella (probably forms of *T. loricatum*) in blood fixed immediately after withdrawal. A single small rounded parasite (6μ in diameter) was seen in our series of slides of freshly-drawn blood. With the exception noted above, every other type of parasite occurring in the developmental cycle described in this paper was observed in fresh-drawn blood. From these observations it seems that this cycle of " swarm " division may be completed in the amphibian host, but that the smallest forms are rarely present in the peripheral blood. It is noteworthy in this connection that there is no certain correspondence between the length of time blood containing *T. loricatum* has been kept and the type of trypanosomes present in it.

More than once large specimens of *T. loricatum* were seen in whose substance red blood cells occurred. Marchoux and Salimbeni believe that the cells and the parasite are merely superimposed. Without wishing to assert that the cell has been ingested, that is, that its presence in the parasite is due to more than a mechanical accident, we are confident that we have seen instances where the cell was definitely within the cytoplasm of the trypanosome. The trypanosomes may be attacked at any stage of their development by leucocytes. Frequently they seem to resist successfully and are not ingested. In one instance a leucocyte was seen to ingest the haemoglobin containing stroma of a disintegrated red cell; it was interesting to note that its protoplasm contained dark brown granules —a most unusual occurrence.

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*Drepanidia.**

Fresh Preparations.

Drepanidia were present in every species of frog and toad named on page 307, and in almost every frog examined. In the same frog (*Rana galamensis*) in which the developmental changes in *T. loricatum* were observed, drepanidia were present in almost every other red blood corpuscle, though scarcely any young parasites were seen. The red cells were swollen to nearly twice their normal size, the colouring matter had disappeared, and the cells in fresh preparations presented the appearance of a crumpled colourless bag, to one side of which the parasite lay. The nucleus was pushed to the periphery of the cell (Plate XXIX, fig. 86).

A few of these drepanidia were found free in fresh preparations examined immediately after making them, but the number of free parasites increased enormously after half an hour. The drepanidia, after leaving the cells and becoming free, were long and had a round anterior end, *i.e.*, the end in advance during progression (Plate XXVIII, fig. 67). A little way from this end the body tapered gradually to the posterior end, which though narrow, was bluntly rounded off (Plate XXVIII, figs. 68 and 69).

Progression takes place in three ways:—

(1) With the body stretched out by a gliding movement.

(2) By gregarine-like contractions of the protoplasm. This occurs when obstruction is encountered (Plate XXVIII, figs. 70 and 71).

(3) By a sudden jerking backward of the posterior end which has previously curved round to meet the anterior end.

* See footnote to page 303.

After about an hour, agglutination of the free parasites was observed. They joined themselves together by their posterior, sharper, somewhat granular ends. Two, four, or six parasites came together in clumps in this way. Contractions occurred from side to side, causing the parasites to bend. Two days after the fresh preparations were made, the agglutinated groups of drepanidia were observed still, the individuals being rather granular and stumpy and actively motile, but they did not undergo any further change.

Three forms of drepanidia were observed, a small form (Plate XXVIII, fig. 74) which will be described in stained preparations, a medium form (Plates XXVIII and XXIX, figs. 82, 68, 69) (the common form), and a very large form (Plate XXIX, fig. 86). These large drepanidia often contained large yellowish, highly refractile granules, which varied in size and number in different parasites. There were from one to about one hundred of these globules in each parasite (Plate XXIX, figs. 84, 85, 89, 90, 91, 92). These highly refractile granules were situated for the most part around the nucleus. In some of these parasites small dancing granules were seen at the more slender end.

Stained Specimens.

The following description is based on the examination of dried and stained films.

The nucleus of the host-cell was displaced (Plate XXVIII, fig. 59) but not injured; the cytoplasm, however, suffers severely since the parasite seems literally to tunnel about within the limits of the cell. Many corpuscles were found in which the contents seemed to have been devoured, leaving them looking like wrinkled empty sacks (Plate XXVIII, fig. 67). Such sacks could be seen with the drepanidia present, or leaving, or gone. Two and three parasites were sometimes seen in one blood corpuscle. The drepanidia were not only present in erythrocytes but were also occasionally found in leucocytes.

As already noted there were three principal forms of drepanidia; a large form, a medium form, and a slender form. The medium form was the most common (Plate XXIX, figs. 81, 82), and was from 15 to 18μ long and from 5 to 6μ wide. It had a fine granular striated protoplasm, which sometimes contained coarse red granules, and sometimes not. Some specimens had a few (four) granules at the anterior end of the body, and some had a straight blue line running

from the nucleus to the anterior extremity. The nucleus was from 3·3 to 4·4μ in diameter, and was placed about 7μ from the anterior extremity and 4μ from the posterior extremity.

One and two constrictions in these forms were observed, showing the method of progression described in fresh specimens (Plate XXVIII, figs. 70, 71). In a few specimens the nucleus had apparently divided into two, and the halves were attached by a red line (Plate XXVIII, figs. 68, 69).

Some of these forms were encysted, both in erythrocytes and free in the serum. Such encysted forms often had a small amount of red-staining excretion at both ends (Plate XXVIII, figs. 59, 60).

The "slender" forms (Plate XXVIII, figs. 73, 74) were from 13 to 15μ long and 1·1μ wide. The cytoplasm was light-staining and of a loose texture. The shorter or posterior end was very faint-staining and the extremity could be seen with difficulty. The longer or anterior end was a faint pink, which deepened towards the extremity. On both sides of the nucleus were areas staining the same as the cytoplasm of the erythrocytes. There were two, sometimes three, of these areas which were from 0·6 to 1·7μ long. Near the posterior extremity there were sometimes from two to four or more red chromatin granules, and occasionally there were a few in the anterior part of the body. The nucleus consisted of eight peripherally arranged chromatic granules, "chromosomes,"* and one or two darker, central chromatic granules, "karyosomes," all connected by fine lines. In one instance (Plate XXVIII, fig. 73) a blue spiral line was found to run from one of the chromatic granules to the anterior extremity of the body. The nucleus was 2·2μ long and was placed 9μ from the anterior extremity, and 3·3μ from the posterior extremity. This "slender" form may be present in the blood with the larger form, or it may be present in blood which has none of the larger forms.

These "slender" drepanidia penetrate the erythrocytes, then lose both ends, so that only the nucleus and a small amount of cytoplasm around it is left. The chromatic granules go to the side, and later they are gathered in a mass at one end of the body, with a few chromatic granules remaining outside the mass (Plate XXIX, fig. 76). The body of the parasite enlarges and at the extremity opposite the chromatin mass are several round clear spaces, which may or may

* It must be understood that the terms chromosomes and karyosomes are used in a purely descriptive sense.

not contain a granule of chromatin. The mass of chromatin divides into two, and the cytoplasm may or may not divide synchronously. In cases where the cytoplasm does not divide synchronously, the chromatin goes on dividing until there are from 10 to 16 masses of chromatin (Plate XXIX, figs. 78-80) arranged almost around the edge of the cell. These rosettes later show divisions into young parasites (Plate XXIX, fig. 79). Rosettes were seen leaving the host-cell, and also found free in the serum (Plate XXVIII, fig. 66). In one slide a group of eleven young drepanidia that had just escaped from the rosette condition was seen (Plate XXVIII, fig. 72). These forms were 7·7μ long and 1·1μ wide. Their cytoplasm was of a faint-staining, coarse, granular structure. The nucleus was composed of a larger, central mass of granules and, usually, a chromatin granule on either side; occasionally both were on one side. In one case the chromatin seemed to be dividing (Plate XXVIII, fig. 72). These sporulating* forms always arose from the "slender" drepanidia. In one series of eight slides from one frog many sporulating and "slender" forms were seen, but none of the larger drepanidia described in the next paragraph.

In a few frogs, *Rana mascariensis*, a very large drepanidium was seen, which was like the common form, but larger. It was lighter stained and often took a pink colour instead of the normal blue. The cytoplasm was coarsely granular, and many of the parasites had non-staining, highly-refractile globules in them. These globules were from one to one hundred in number, and from 0·6μ to 7·7μ in diameter. They were always found near the nucleus and gave the appearance of a fatty degeneration of the nucleus. They sometimes distended the drepanidium until it was nearly round (Plate XXIX, fig. 89), but they usually did not affect the size of the parasite. The large type of drepanidia presented three forms, a long form, a short or young form, and a folded or "two-shanked" form. The long form (Plate XXIX, fig. 86) was usually 22·6μ long and 7μ wide. The nucleus was 3·4μ long and 5·6μ from the posterior extremity and 13·6μ from the anterior extremity.

The short, or young form (Plate XXIX, fig. 83) was about 8·8μ long and 6·6μ wide. The nucleus was 4·4μ in diameter and placed in the centre of the organism.

The folded form was seen in the various stages of unfolding (Plate XXIX, figs. 87, 88).

* The term is not used in its specific sense.

Cytamoeba.

Cytamoebae were present in a few of the frogs in considerable numbers. Amongst the forms seen was the round form (Plate XXIX, figs. 104, 106, 108, 110) previously described in the snake (page 304) (2·2μ wide). In it were short rods, or sometimes round, red dots (Plate XXIX, fig. 104). Other forms were oval or finely granular, sometimes with no apparent internal structure (Plate XXIX, fig. 103). The round forms were often found near the nucleus, and sometimes apparently emerging from it, giving the appearance of fragmentation of the nucleus (Plate XXIX, fig. 110). Some of the parasites were filled with slender rods, which at times projected beyond their edge for a distance of 4μ (Plate XXIX, figs. 112, 113). Other forms were present which looked like masses of rods bound together (Plate XXIX, figs. 107, 108, 109, 115); sometimes these bundles of rods were free in the serum (Plate XXIX, fig. 100). Occasionally short rods were found free in the erythrocytes, sometimes end to end, sometimes crossing one another at right angles. The rods found in all the above forms were of two kinds, a short rod with rounded ends, and a long filamentous rod. Some of the short rods were seen free in the blood plasma in such forms and arrangements that there seems to be no doubt that some of them at least are bacteria. It is equally certain that some of the longer slender rods are merely filamentous cytoplasmic processes of the rounded parasite. In some of the specimens the body of the cell was stained a light blue, while the rods within were stained a bright red.

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An unidentified parasite

Another parasite, a round, red-staining mass from 1·5 to 5·5μ in diameter, is found in red cells (Plate XXIX, figs. 93, 95, 99). With it are associated bluish-green, crystal-like bodies (Plate XXIX, figs. 93, 95, 97, 98). Both the red masses and crystals may be present in varying numbers (Plate XXIX, figs. 98, 99). The cytoplasm of the host-cells is usually stained a deeper blue than usual. Many of the smaller red masses have a well-defined blue area around them, which suggests that this parasite possesses a cytoplasmic body (Plate XXIX, figs. 94, 96). In stained films of fresh blood the red masses do not seem to have much structure, but in films of blood kept for two days, the structure consists of chromatic granules arranged peripherally with one or two darker chromatic granules in the centre.

This parasite with its crystals was seen in fresh specimens. It occurred in conjunction with drepanidia and *T. loricatum*.

FISHES*

Fresh-water fish of many different sorts were examined in the Gambia and in the Congo. Parasites were found only in the fish without scales, mentioned below.

Those infected were caught in a very small sluggish stream with a very dirty bottom; none of them were over seven inches in length and none were infected with drepanidia.

Trypanosomes (Plate XXX).

Two "mudfish" (*Clarias angolensis*) caught at Leopoldville in the Congo Free State on December 9th and December 30th, 1903, had actively motile trypanosomes in their blood which may be divided into three types; a small, a medium, and a long form. The small type was identical in both, the medium was present in both, but with slight differences, and the long form was present only in the fish of December 9th. The small form was very frequently seen in the fish of December 30th, while in the fish of December 9th, only four were present in a total of ninety-nine parasites. The medium form, in the fish of December 30th, was less frequently met with than the small form, but was often seen in the fish of December 9th. Of the long forms only thirteen were seen. All the forms were characterised by the possession of a large four-lobed blepharoplast,

* The description of the parasites found in fishes is republished by permission from the Journal of Medical Research.

situated at, or very near, the posterior extremity, and by a clear space about the nucleus. The blepharoplast apparently consisted of four darkly-staining granules of equal size embedded in a matrix. In a few specimens there were, perhaps, more than four granules in the blepharoplast, but in these cases it was never possible to be certain of the exact number. The three forms of parasites seemed but different variations of one species, as gradations between all forms could be seen.

The small type was readily distinguished by its size, its narrow body, its relatively long nucleus—situated more anteriorly than in the other two types, and with its long diameter parallel to that of the body—and by its faint-staining reaction. The blepharoplast was oval, or round, large and distinct (measuring 1·13 by 0·75μ), and was situated at the posterior extremity. The undulating membrane was well developed, having a width of 0·75μ. The long, oval nucleus was situated at the junction of the middle and anterior thirds of the body. It was granular in structure and contained a fibrous network on which were seen eight to sixteen chromatin granules, and one darker granule—the karyosome. The body protoplasm was reticular, and contained a few violet-staining granules. Occasionally parasites were seen in which the body was filled with these coarse chromatophilic granules. Superficial longitudinal striations to the number of five could be seen near the nucleus of two slightly injured parasites. Division forms were very numerous in this type (even when but four were seen in the fish of December 9th, one of them was dividing). They followed the usual method of longitudinal division. In the division of the blepharoplast, the posterior lobe on the concave side of the parasite moved behind the other posterior lobe; the axis of the posterior lobes thus became longitudinal, while that of the anterior lobes remained transverse. The two posterior lobes then separated from the anterior lobes and their axis became transverse again.

The medium type was characterised by its size, and its darker staining reaction. The blepharoplast was large, oval or round, distinct and measured about 1·5 by 0·75μ. It was situated at the posterior extremity, or from 1 to 1·5μ anterior to it. The flagellum was relatively longer than in the short form. The undulating membrane in parasites from the fish of December 30th was well

developed, and wound in numerous folds around the body; it was 1·5 μ wide. In trypanosomes from the fish of December 9th, the undulating membrane was poorly developed, had a few folds and was not more than 1 μ in width. The nucleus was round to oval, its long diameter was at right angles to that of the body, and it occupied the whole width of the parasites. It was granular in structure and contained a fibrous network on which were seen sixteen to twenty-four chromatin granules and one or two darker granules—the karyosomes. The body was more darkly stained in the fish of December 30th. Superficial longitudinal striations to the number of seven were counted near the nucleus. Divisional forms were few in this type and followed the usual method of longitudinal division.

The long type was characterised by its size, its tapering extremities, its comparatively short flagellum and by the possession of from one to four vacuoles just anterior to the blepharoplast. The blepharoplast was large, oval or round, and situated 2 μ from the posterior extremity; its measurements were the same as those of the medium type. The nucleus had the same characteristics as that of the middle type, except that it did not quite occupy the width of the body. The protoplasm was the same as in the medium type, except for the above mentioned vacuoles, which were always present. Eight superficial longitudinal striations were present and these were seen to cross the nucleus. No divisional forms of this type were seen.

The measurements of the three types were as follows:—

	Small form	Medium form	Long form
Measurement I	... 0·0 μ	... 0·0 to 1·5 μ	... 2·0 μ
,, II	... 12·7 μ	... 14·2 μ	... 25·0 μ
,, III	... 3·0 μ	... 3·7 μ	... 45·0 μ
,, IV	... 6·7 μ	... 12·0 μ	... 19·5 μ
,, V	... 12·0 μ	... 13·5 μ	... 10·5 μ
,, VI	... 1·0 μ	... 3·0 μ	... 4·5 μ
Total length ...	... 34·5 μ	... 45·0 μ	... 61·5 μ

Although the trypanosome described by Montel[1] closely resembles our large form, we have seen no account in the literature of any parasite morphologically identical with those described above.

LITERATURE

Good lists of the publications on the trypanosomata of fishes will be found in the works of Laveran and Mesnil, "Trypanosomes et Trypanosomiasis," and of Mense, "Handbuch der Tropenkrankheiten."

The following are important papers not mentioned in these volumes:—

1. MONTEL. Trypanosoma d'un poisson de Cochinchine. Comptes Rendus de la Société de Biologie, T. I, 1905, p. 1016.
2. LEBAILLY. Recherches sur les Hématozoaires des Téléostéens marins. Thèse Fac. Med. Paris, 1906, p. 76; and Archiv. de Parasitologie, T. X, 1906.
3. LEBAILLY, M.C. *Trypanosoma bothi* (Barbue) n. sp. Comptes Rendus de la Société de Biologie, T. IX, 1905, p. 304.
4. BRUMPT. Sur quelques espèces nouvelles de Trypanosomes parasites des Poissons d'eau douce. Comptes Rendus de la Société Biologie, T. LX, 27th Jan., 1906, page 160.
5. BRUMPT. Mode de transmission et évolution des Trypanosomes parasites des poissons. Comptes Rendus de la Société de Biologie, 17th Jan., 1906, T. LX. No. 4, p. 162.
6. BALFOUR. Second Report of the Wellcome Research Laboratories, Khartoum, page 197.

A Spirochæte (Plate XXXI).

In the blood of the fish of December 30th a spirochaete-like organism was found which could be identified with no previously described spirochaete.

It had a hair-like body; its length was usually 18μ; its breadth at the widest part was 0·6μ. It was widest at one extremity or towards the middle; in the latter case, the parasite tapered towards both ends, but one extremity was always much thicker than the other. The wider end was rounded, while the other end was drawn out to a fine and slender point. The parasite usually occurred singly and lay in a simple curve, forming three-quarters of the circumference of a circle (Plate XXXI, figs. 1 and 2). Other forms—coils (fig. 3), compound curves (figs. 4, 9-12) and spirals (figs. 5-8) were seen as illustrated. The coils were always formed by the slender end of the parasite. The spiral forms were long, measuring from 24-27μ, and in some of these forms there was a suggestion of a membrane (figs. 5 and 6). The parasites were also seen in pairs and in groups of from three to many individuals as shown in figures 13, 14-16. These organisms seemed to consist of a deeply-staining core and a lightly-staining periplast. Many of them stained irregularly by modifications of Romanowsky's method. The lighter stained areas (fig. 2) occurred at irregular intervals and were of

irregular extent. The periplast could be seen at these lightly stained areas and it was continued to form the slender extremity from which the core was also absent. Occasionally granular forms were seen in which the protoplasm of the core apparently arranged itself into small granular masses, but no regularity could be detected in the arrangement of these granules. Undoubted multiplication forms were not seen.

For descriptive purposes we propose for this organism the name of *Spirochæta jonesii.*

ARTHROPODA

The protozoa found in this class were encountered by chance during an investigation of the development of various haematozoa, while examining various blood-suckers, either as controls, or for the presence of developmental forms of the blood parasites. Many tsetse flies and mosquitoes, a few *Stomoxys*, ticks of various sorts, fleas and lice, and "Congo floor maggots," were examined. The only protozoan parasites observed are recorded below.

MOSQUITOES

(1) *Myzorhynchus paludis.*

A cluster of radiating club-shaped bodies (Colonie radiée) was seen in the thoracic tissues of a female of the above species, examined for malaria at Lusambo. The parasites became free, developed a flagellum and were actively motile in the salt solution in which the mosquito was dissected.

(2) *Pyretophorus costalis.*

Some excitement was at first created during the examination of the alimentary canal of mosquitoes, fed at varying antecedent periods on animals infected with trypanosomes, by the presence of vermicule-like bodies (Plate XXXII, fig. 14), measuring about 18 by 2·2μ. These parasites progressed slowly by active lashing and slower amoeboid movement; some similar forms were motionless.

The amoeboid movements were of two sorts. The first involved a change in the whole shape of the parasite. The second was by a simple protrusion of protoplasm from the rounded anterior, never from the effilated posterior extremity.

They occurred either within the stomach, in the stomachal tissues, or in the body cavity of the mosquito. A central differentiated area could be detected in them by examination in fresh specimens; near the area were more or less numerous granules which often exhibited lively Brownian movement. After some time the parasites became much stumpier (8μ by 16μ), and were later almost spherical. One parasite, however, remained unchanged and active for twenty-four hours.

Further search showed that the parasite occurred in freshly-caught and freshly-hatched mosquitoes, and in the larvae from which all our experimental mosquitoes were raised, and it was quickly shown to be a stage in the life cycle of a coccidium. It was most interesting that practically every larva from one pool was infected with this coccidium, while none of the larvae and adults (anopheline and culicine) from a pool only a few yards distant were affected. So far as was observed the parasite did not occasion any excessive mortality amongst the mosquitoes infected. This seems difficult to understand—the mosquitoes were probably not watched long enough—since the tissues of many of them were fairly riddled with coccidium cysts. The water of the heavily-infected pool was centrifugalised and examined. Coccidium cysts were not recognised, various infusoria and a clump of herpetomonas-like flagellates were alone seen.

So soon as it was certain that the vermicule-like "sporozoite" had no connection with the trypanosomes ingested, the study of this coccidium was discontinued. It is, therefore, not possible to say whether the forms observed represent stages in more than one parasite or no. It seems probable that but one species of coccidium was present. We describe the forms observed and indicate the position they seem to occupy in the life cycle of the parasite. The mobile sporozoites first seen become free through the rupture of a sporoblast containing eight sporozoites. The number of sporozoites in each sporoblast is almost invariable. Fig. 15 is unusual in that the nucleus of two of the sporozoites has divided, and in one instance division of the cytoplasm seems to have commenced. As a rule the long diameters of the sporozoites are parallel. These are, therefore, regularly arranged like the segments of an orange (corps en barillet). Single sporoblasts surrounded by a definite cyst wall may occur. They are more usually seen in groups of four, seven, or even as many

as eleven sporoblasts packed together in an oocyst with a definite capsule. The individual sporozoites have no capsule. Their cytoplasm stains a light blue by Romanowsky's method. It is alveolar in structure and rarely contains granules of any sort, but may have occasional granules. The nucleus is placed centrally, is loose in texture and consists of a varying number of chromatin granules placed in a more lightly-staining matrix. From the analogy of other coccidia, we assume that the forms just described represent the completion of sporogony. The male and female gametes, which probably commence this cycle, and their conjugation have not been seen. Neither has the further development of the sporozoites been observed.

The number of rounded merozoites (figs. 16, 17, 18), present in every part of the larvae and adults dissected, was often extraordinary. These organisms measured about 4 to 5μ in diameter. They were either free or enclosed in a host cell. On becoming extracellular they were seen to possess a definite capsule (pink-staining); this was quickly discarded (figs. 18, 19) and the empty shells were very frequently seen. Often merozoites which had just cast their capsules occurred in groups of four (fig. 18). Four merozoites were, however, never seen within an unbroken capsule. As a rule whether intra or extra-corpuscular the merozoites occurred singly. When extra-corpuscular they were occasionally in pairs. Sometimes large intra-corpuscular cysts were seen which contained very large numbers of merozoites. The merozoites (figs. 16, 17, 18) consisted of a pinkish alveolar cytoplasm in which was situated a nucleus usually surrounded by an indefinite bluish area. In this bluish area a lighter, rounded spot was almost invariably present. A few chromatophilic granules and small clear spaces were frequently seen in the cytoplasm. The nucleus consisted of irregular masses of chromatin placed in a matrix. In some forms (fig. 16) the granules were placed at the periphery of the nucleus and were joined by threads with a central darker-staining mass. In fresh specimens they were immobile.

LITERATURE

1. Ross. Parasites of Mosquitoes. Jour. of Hygiene, Vol. VI, No. 2, April 1906. The subject is reviewed and discussed.

TSETSE FLIES (*Glossina palpalis*)

Parasites of the type described below were found in specimens of *Glossina palpalis* freshly-caught, and at all periods, up to 11 days, after having fed on known animals infected with trypanosomes. All the parasites were not precisely similar, but one description will suffice for all. The free rod-like parasites were found only in the alimentary canal, nearly all of them in its anterior part. They were most numerous near the oesophagus, in the "stomach" or in the neighbouring part of the intestine. Cysts containing them were seen attached to, or actually in, the wall of the intestine. Some of them were whole, others had burst (or were burst by slight pressure), and from them were expelled the rod-like bodies. In fresh preparations these rods (Plate XXXII, figs. 1, 2, 3, 4, 5, 8, 9, 10) were non-motile, they contained one or two refractile granules and they occurred singly or, occasionally, in small clumps. The most usual form seen in stained specimens was a rod about 7·0μ in length and 1·0μ in width with abruptly rounded ends (figs. 1, 2, 5). It was limited by a definite outline, but apparently not by a capsule. Its body substance consisted of pale blue protoplasm in which occurred chromatophilic granules and rounded clear areas. The chromatophilic granules varied in size from extremely fine particles (fig. 6) up to masses measuring about 0·3 to 0·5μ in width (fig. 3). They were usually distributed more or less regularly in transverse bands (figs. 1, 2, 5, 10) lying, at intervals, across the parasite. In such parasites the individual granules could barely be distinguished. Often between each band of granules was placed one of the rounded, clear areas (figs. 1, 2, 5, 10). Such parasites were probably preparing for transverse division, as small forms, apparently consisting of one or two rounded segments of the original rod-like body, were often observed (fig. 7). In some of the parasites showing the band-like distribution of chromatin, as well as in others in which pairs of granules were arranged in bilateral symmetry (figs. 8, 9), it seemed as though longitudinal division might be about to occur. In many parasites the chromatin granules were distributed absolutely without order (figs. 3, 4, 6). Rarely a larger and a smaller granule seemed to be in connection with each other.

Rod-shaped parasites of all these three types were frequently terminated by a small chromatophilic granule (figs. 4, 5, 10) (sometimes placed in a clear area), or by a rounded enlargement which

often measured twice the diameter of the rest of the parasite (fig. 10). Rounded parasites measuring from 3μ to 5μ or more in diameter with irregularly distributed granules and no clear areas occurred (figs. 11 and 13). The smaller rounded forms can divide transversely; apparently after a concentration of the chromatin (fig. 12).

It seemed possible that this parasite should be placed amongst the *Myxosporidia*. In recognition of the collaboration of Dr. Inge Heiberg in the work of our expedition, and in particular of his observations on *T. loricatum*, we suggest for the name of this parasite *Myxosporidium heibergi*. About half of the tsetses dissected at Leopoldville had these parasites free in their midgut. In the stomach wall of one freshly caught fly coccidian-like cysts were seen. (" Corps en barillet " ; compare the parasite of *Pyretophorus costalis* described above).*

EPILOGUE

Although the facts presented in this paper are admittedly the results of passing observations, incomplete, disconnected, and made under unfavourable circumstances, we think that they possess a certain value as a record alone of protozoan infections encountered by chance during three years' work in Africa. Many of the appearances noted, though incomplete in themselves, are of additional value in that they confirm the constancy of morphological changes noted in other allied and better studied parasites.

The following examples are cited. Longitudinal striations have been seen in every trypanosome examined; the myonemes of other authors are, therefore, constant in the trypanosomes we have dealt with. In several instances the thickened edge of the undulating membrane of trypanosomes has been observed to terminate, not in the blepharoplast, but in a lighter-staining pinkish granule, or pair of granules, closely applied to it. In some of our trypanosomes the structure of the nucleus and the occurrence, in neighbouring differentiated areas, of paired chromatophilic granules suggests a connection with the complicated nuclear developmental phenomena observed in trypanosomes by some observers. The clear, unstained

* This is the last of the parasitic protozoa observed during our expedition to the Gambia and the Congo. The filariae found in various mammals, birds, amphibians and reptiles will be considered in a forthcoming paper by Professor. H. E. Annett.

perinuclear area, the irregular, non-staining almost granular areas, and the lines in the cytoplasm as well as the chromatophilic granules (see Plate XXVII, fig. 34) frequently observed in various trypanosomes described in this paper, are all constant phenomena, which seem to be of biological significance. In the development of *T. loricatum* the work of Danielewsky was confirmed and extended to prove a most interesting cycle of multiplication in a well-known trypanosome; it must be asked whether a similar cycle may not occur in other trypanosomes. The rough resemblances between the spirochaetes seen in ulcers and in fish with previously described parasites of this nature are striking. In the leucocytozoon of birds an interesting course of development is described.* Unfortunately our material was too scanty to permit us to fully compare our observations with those of Schaudinn. Very little has been said concerning the drepanidia observed. In spite of a very careful search in kept preparations of blood, we were unable to find any indication of a direct relation between this parasite and the trypanosomes which were often associated with it.† Neither were parasites resembling *Trypanosoma inopinatum* seen to become intracorpuscular.‡ In both stained and fresh preparations of frogs blood, however, the great resemblance in size and general appearance between some trypanosomes and some drepanidia was very apparent. It was also noticed that, as a rule, if there were many drepanidia in a given frog, there were also many trypanosomes.

One of the earliest of the lessons learned from our work on these protozoa was the entire inadequacy of the methods of preparing blood-films, ordinarily employed by pathologists, for a morphological study of protozoa.

The examination of dried films stained by modifications§ of Romanowsky's method suffices for the purposes of clinical diagnosis. This method will show many of the structures present, it was used in

* The dangers of constructing a part of the life-history of a parasite from stained specimens alone are apparent. The course of development we described may be mistaken, but the descriptions of the forms mentioned are accurate and may be relied upon so far as the defects of the method of preparation employed permit.

† Billet, Culture d'un trypanosome de la grenouille chez une Hirudinée. C. R. Acad. des Sci., T. CXXXIX, Oct. 10, 1904, p. 643.

‡ Billet, *Trypanosoma inopinatum* et sa relation avec les Drepanidium. C. R. Soc. de Biol., July 23, 1904, p. 161.

§ Stephens and Christophers. The Practical Study of Malaria, page 10, University Press of Liverpool.

the whole of the work described in the present papers, but it fails to demonstrate finer details properly. Our work must, therefore, be regarded as incomplete, and certainly the more important parts of it should be repeated by workers using methods more perfect than those employed by us.

A second lesson was that too much of the work done on the pathogenic protozoa, particularly by medical men, has been directed by conceptions derived from bacteriology.

Days of tedious searching of slides and of observation of parasites, placed under various conditions, must be spent in studying the pathogenic protozoa, when minutes would almost suffice in the case of bacteria. The difficulty of finding the parasites at all is sometimes extraordinary, and the possible occurrence of a latent infection must never be forgotten.

The study of the pathogenic protozoa must be approached with an unbiassed mind and with the remembrance that the known life-cycles of several protozoa are exceedingly complicated. We believe that the continuous observation of living parasites will ultimately furnish the richest reward. A single positive observation so obtained is absolute, and outweighs any number of apparently antagonistic probabilities obtained by deductions from work done along apparently parallel lines of research. Of course, the examination of fresh preparations should be supplemented by the examination of stained material. Lastly, at the present moment more is known, in every way, of malaria than of almost any other disease. In observing less studied protozoan infections it will frequently happen that our knowledge of what actually does occur in malaria will lead to the formation of an ultimately successful working hypothesis, or to the correct interpretation of newly-observed phenomena.

EXPLANATION OF PLATES

All the illustrations accompanying this paper, except where it is otherwise stated, are drawn to a magnification of 2,000 diameters. No camera lucida was employed; the dimensions were obtained by measurement. A Zeiss 2mm. apochromatic 1·40 aperture objective, with an 8 or 18 eyepiece, was regularly used.

PLATE XX

Figs. 1 to 28, except 27, are "Young Forms"

As a rule, ectoplasm, endoplasm and nuclear material can be distinguished in each of these parasites; occasionally it is difficult to do so (fig. 24). The arrangement of the parasites and the host-cell are well seen in figs. 1, 25, 28. The constant presence of a clearer area about the chromatic material is to be noted in these parasites.

Fig. 1.—Chromatic mass with linear extension possibly antecedent to nuclear division, as in fig. 3.

Fig. 2—Shows nucleus with vacuole containing indistinct granules.

Fig. 3.—Nuclear material divided into a small and a larger chromatin mass connected by a line; possibly stage succeeding fig. 1.

Figs. 4, 5 and 9.—Chromatin divided into a larger mass and a smaller granular one, placed in a vacuole. Note the vacuole in these parasites is frequently pink-staining.

Figs. 6 and 7.—Forms similar to 2 and 5 in very small parasites.

Fig. 8.—Very small parasite showing a line.

Fig. 10.—Chromatin divided into two equal masses.

Figs. 11 and 12.—Are squashed and somewhat degenerated parasites. They are useful in that they show distinctly the presence of definite structure in the nuclear material.

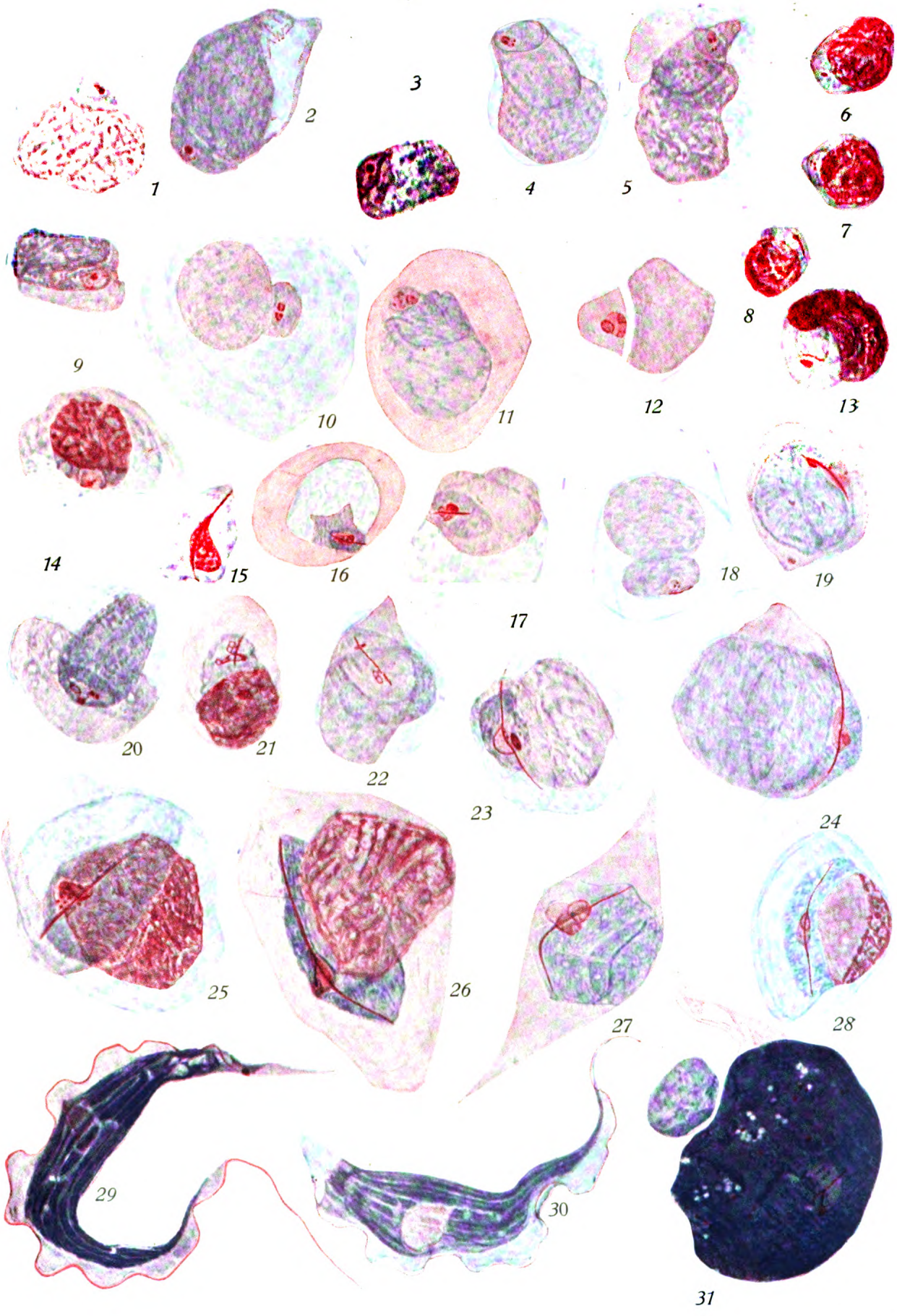

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Fig. 13.—Shows a form in a small parasite probably comparable to 9. The vacuole is well marked. The endoplasm of the parasite has pressed upon and considerably distorted the nucleus of the host-cell.

Fig. 14.—Probably not similar to 1, but commencing formation of line.

Figs. 16, 17 and 18.—Early formation of line. Note its position in the chlamydoplasm.

Figs. 19, 20, 21 and 22.—Are unique specimens. They probably represent important stages of the maturation of the parasites. We feel it unwise to attempt to discuss their exact significance. In 22 a wavy bluish-line is disposed along "the line."

Fig. 15.—Is a squashed and degenerated parasite. The immediate connection of at least one pair of chromatophilic granules with the line is very evident in 15, 19 and 23.

Figs. 23, 24, 25 and 28.—Are well-grown parasites which retain their spherical form.

Fig. 26.—Shows commencing transverse division of the line. The connection of the "blepharoplast" with the blunt extremity of one half of the line is well shown.

Fig. 27.—Line shows commencing longitudinal division. Note that there are two distinct areas of chromatophilic material, beside the chlamydoplasm, lying immediately beneath the dividing line. (This specimen was unique.)

Figs. 29 and 30.—Trypanosomes.

Fig. 31.—Spherical female. Note dark granule connected with line running across chlamydoplasm; this is the only instance in which such a line was seen. The ectoplasm and host-cell nucleus are being thrown off.

PLATE XXI

With the exception of fig. 38, probably a female, all the parasites illustrated in this plate are males.

Fig. 33.—Division of the line; nucleus only just commencing to divide.

Fig. 33a.—Division of line with chlamydoplasm and nucleus. The ectoplasm has been deleted for convenience of reproduction.

Fig. 34.— Formation of line (?). Note the presence of granules of chromatin.

Fig. 35.—A very common form (see figs. 42, 43, 74). It represents the line passing through a diffuse chromatic area (chlamydoplasm) in which lies a denser chromatic area (nucleus ?), often containing one or more (figs. 42, 41) deeply chromatic granules (blepharoplast ?). Occasionally these granules, or similar ones, lie outside the denser nuclear material (fig. 43).

Fig. 36.—Degenerated and squashed parasite, showing multiple nature of line.

Fig. 37.—Only three parasites of this type (all practically identical) were seen. It is remarkable in possessing a blepharoplast-like mass of chromatin distinct from the main nuclear structure. There is an indefinite arrangement of fibrillae and granules about the nucleus and chlamydoplasm.

Fig. 38.—A female cell (unique), in which a faint blue line was wound spirally about one half of the line. Appearances resembling this were seen in one or two other preparations (fig. 62).

Fig. 39.—Line dividing longitudinally, nucleus already completely divided. Parasites of a similar type are not infrequent. The one illustrated is considerably distorted.

Figs. 40 and 41.—Commencing transverse division of the line. In each parasite the line has become thinner. In fig. 41 the differentiated area surrounding the constriction is well seen.

Figs. 42, 43 and 74.—Are ordinary types of adult males with lines.

Figs. 41, 42 and 43.—Blepharoplastic (?) granules occur in the two former within, in the latter without, the nucleus. The presence in fig. 43 of a bluish area within the chlamydoplasm in immediate connection with the nucleus is suggestive.

Fig. 44.—The line is divided transversely (specimen unique); the two halves are connected by a filament.

Fig. 45.—(Compare figs. 63 and 64). Probably represents a stage anterior to fig. 46. (Specimen unique.)

Fig. 46.—The line is much involuted. It extends far into the ectoplasm. A faint blue spiral line (artefact ?) extends, from a double granule in the ectoplasm, through the lower part of the parasite. The arrangement of the nucleus is not at all definite.

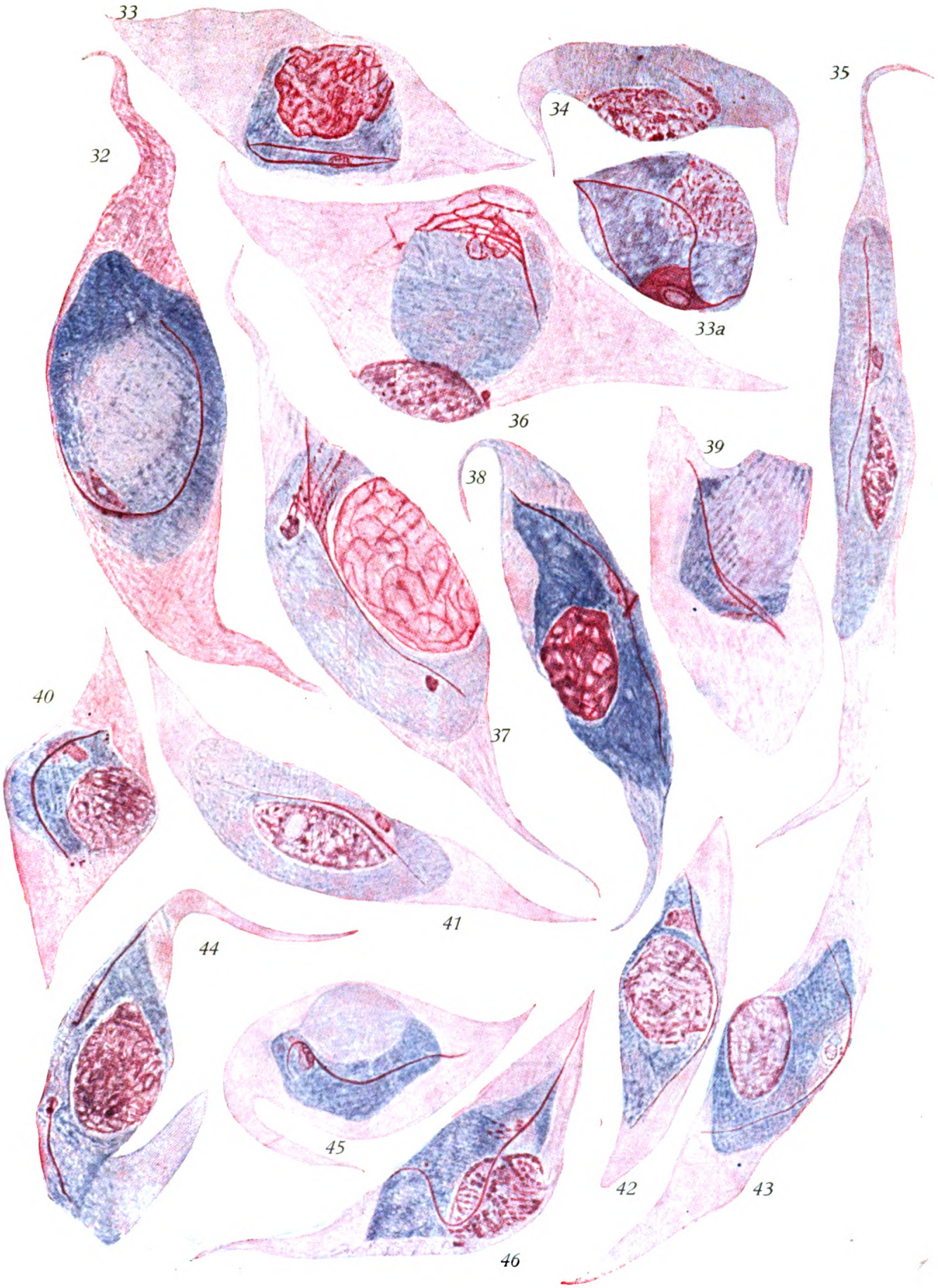


PLATE XXI.

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PLATE XXII

Fig. 47.—(Specimen unique. The colouring of this parasite is much too vivid.) Probably occurs at almost the same period as fig. 49. It is remarkable for the four granules placed in the concavity of the crescentic nucleus. (Similar dots have been seen in other parasites, but never again in this position).

Figs. 48, 49.—Are very common types of males. Note the spherical nucleus with its neighbouring granules in 48 and the distributed chlamydoplasm in fig. 49. (Compare fig. 75.)

Fig. 50.—Shows a disjointed bit of chlamydoplasm in relation with the blepharoplast. Note the granules in this position. The alveolar structure of the endoplasm has given the parasite a lattice-like appearance.

Figs. 51, 52, 53 and 55.—Are stages in the division of the nucleus. In fig. 53 the vacuole in association with the blepharoplast is well seen.

Figs. 54, 56 and 59.—The chlamydoplasm is abundant. The nucleus has formed a spindle-like figure, best seen in fig. 56. In fig. 59 the chromatic granules of the figure seem about to divide, and the vacuole in connection with the blepharoplast is well seen.

Fig. 57.—Two tiny granules are attached to the blepharoplast by fine threads. The nuclear chromatin is commencing to show polar concentration. (See fig. 60.)

Fig. 58.—Granules are being extruded (?) from both nucleus and blepharoplast.

Fig. 60.—Is probably the stage succeeding fig. 57. The nucleus has almost divided.

Fig. 61.—Shows enormous increase of chlamydoplasm. (Compare fig. 56.)

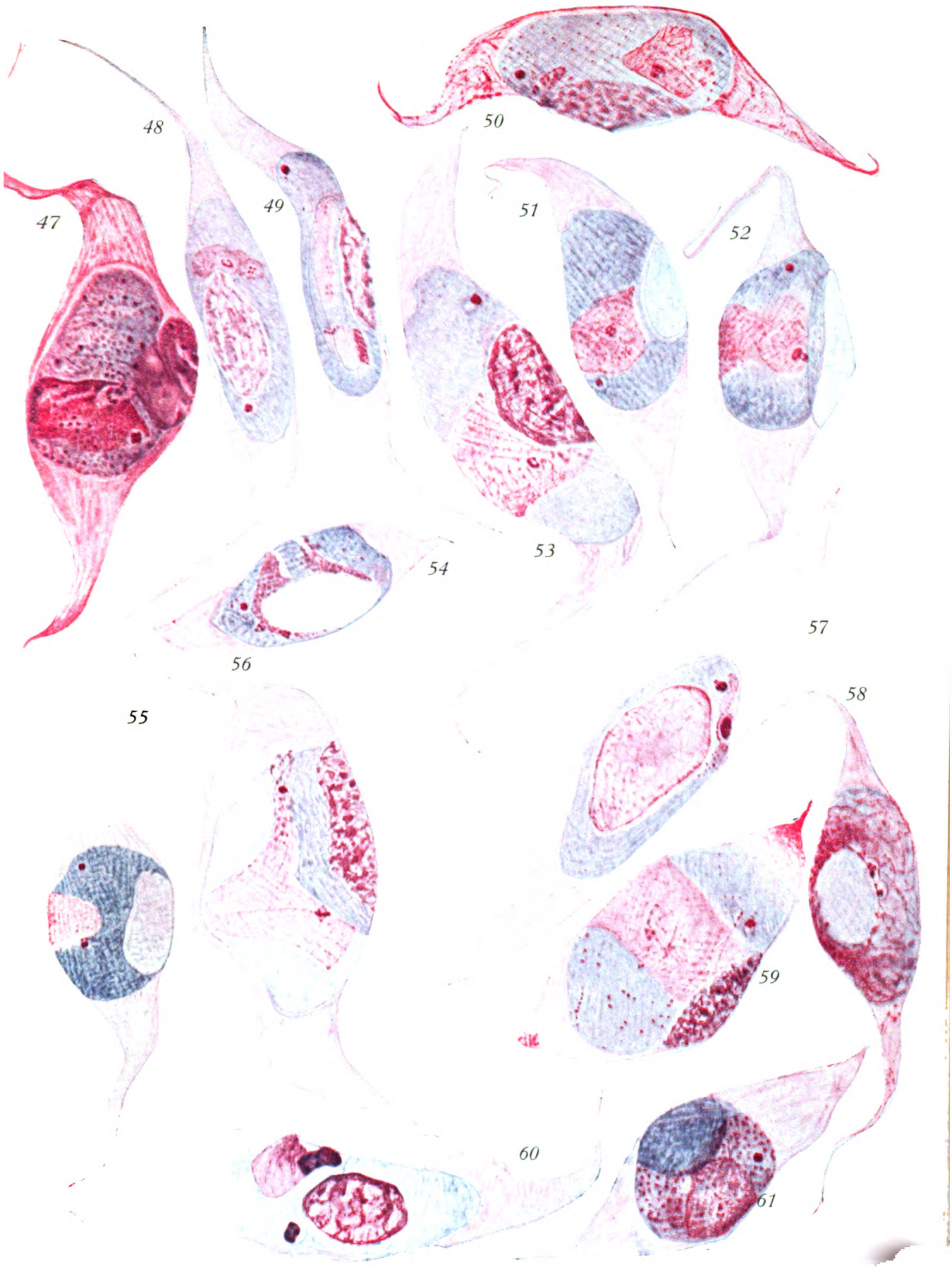


PLATE XXII.

PLATE XXIII

× 1,334.

Figs. 62-73, except 68, 70, and 71 are females.

Fig. 62.—Is a stage in line formation. Note the wavy blue line crossing the chlamydoplasm.

Figs. 63 and 64.—Compare fig. 45.

Fig. 65.—Note endoplasm lying over host-cell nucleus. Chlamydoplasm does not stain, so nucleus appears to lie in a vacuole.

Fig. 66.—Host-cell nucleus extruded, nucleus well differentiated. Note granules in chlamydoplasm.

Fig. 67.—Host-cell nucleus about to be extruded. Compact chlamydoplasm, well-defined nucleus (or blepharoplast?). This is a very common type.

Fig. 68.—(Compare figs. 48, 57, 71, 75.) Chlamydoplasm with spherical nuclear area. Blepharoplast very granular and one granule extruded. (Compare fig. 49.)

Fig. 69.—(Compare fig. 67.) Is a common type. The chlamydoplasm is very diffuse.

Fig. 70.—Parasite possessing distinctive characters of neither adult male nor female.

Fig. 71.—Note deeper-staining area in nucleus. (Compare fig. 68.)

Fig. 72.—Much effilated makrogametocyte; in one instance two fine chromatic granules occurred, as illustrated, in the chlamydoplasm.

Fig. 73.—Is a common type of adult female; the vacuole in connection with the blepharoplast is well seen.

Fig. 74.—Compare fig. 35.

Fig. 75.—(Compare fig. 48, &c.) Note the two large ill-defined granules outside the nucleus in the chlamydoplasm.

Fig. 76.—(Compare fig. 49.) Is palely stained; chlamydoplasm is much extended.

Fig. 77.—Discarded ectoplasmic sheath and host-cell nucleus of an adult parasite.

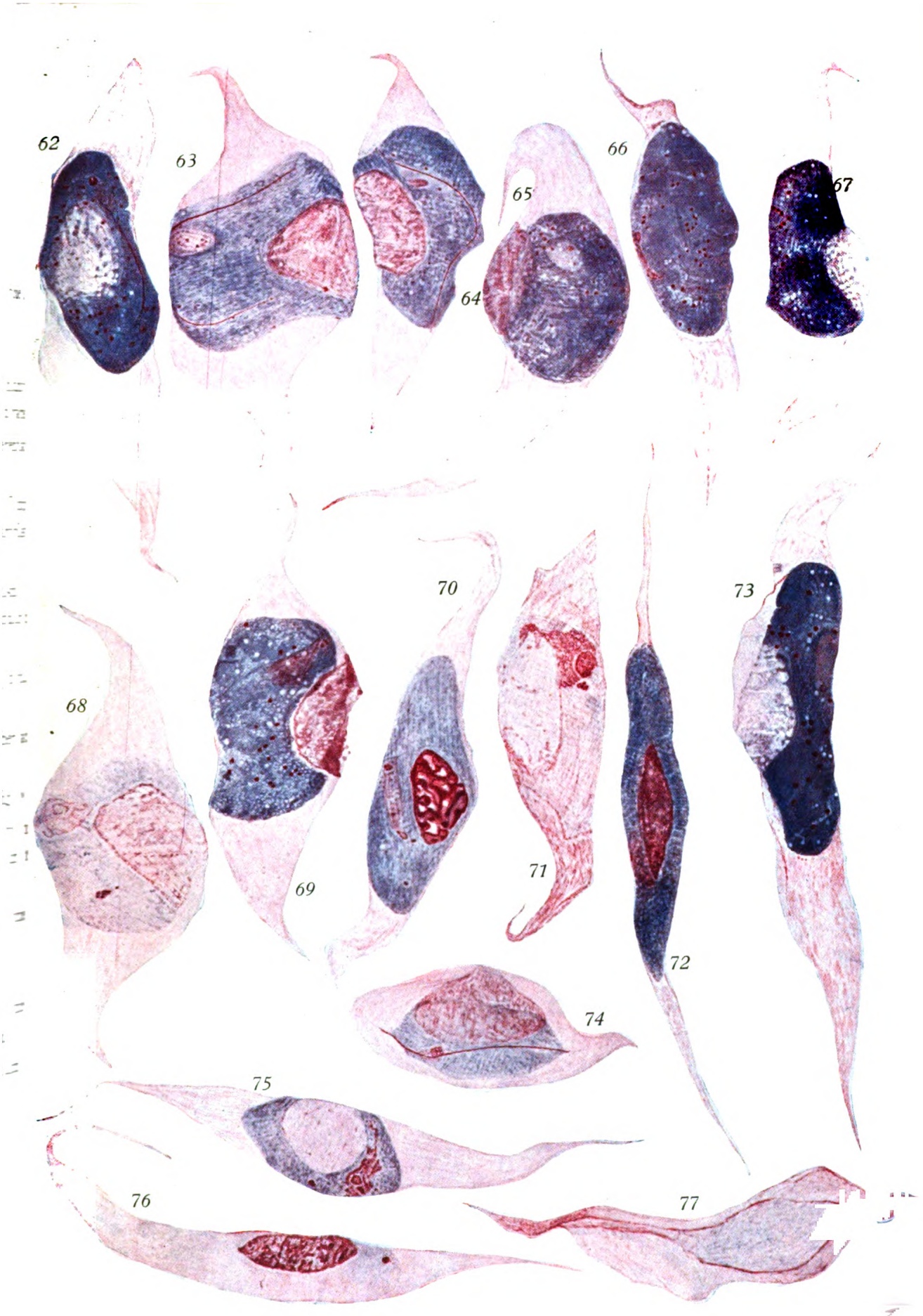


PLATE XXIII.

PLATE XXIV

Fig. 1.—" Slender " form.

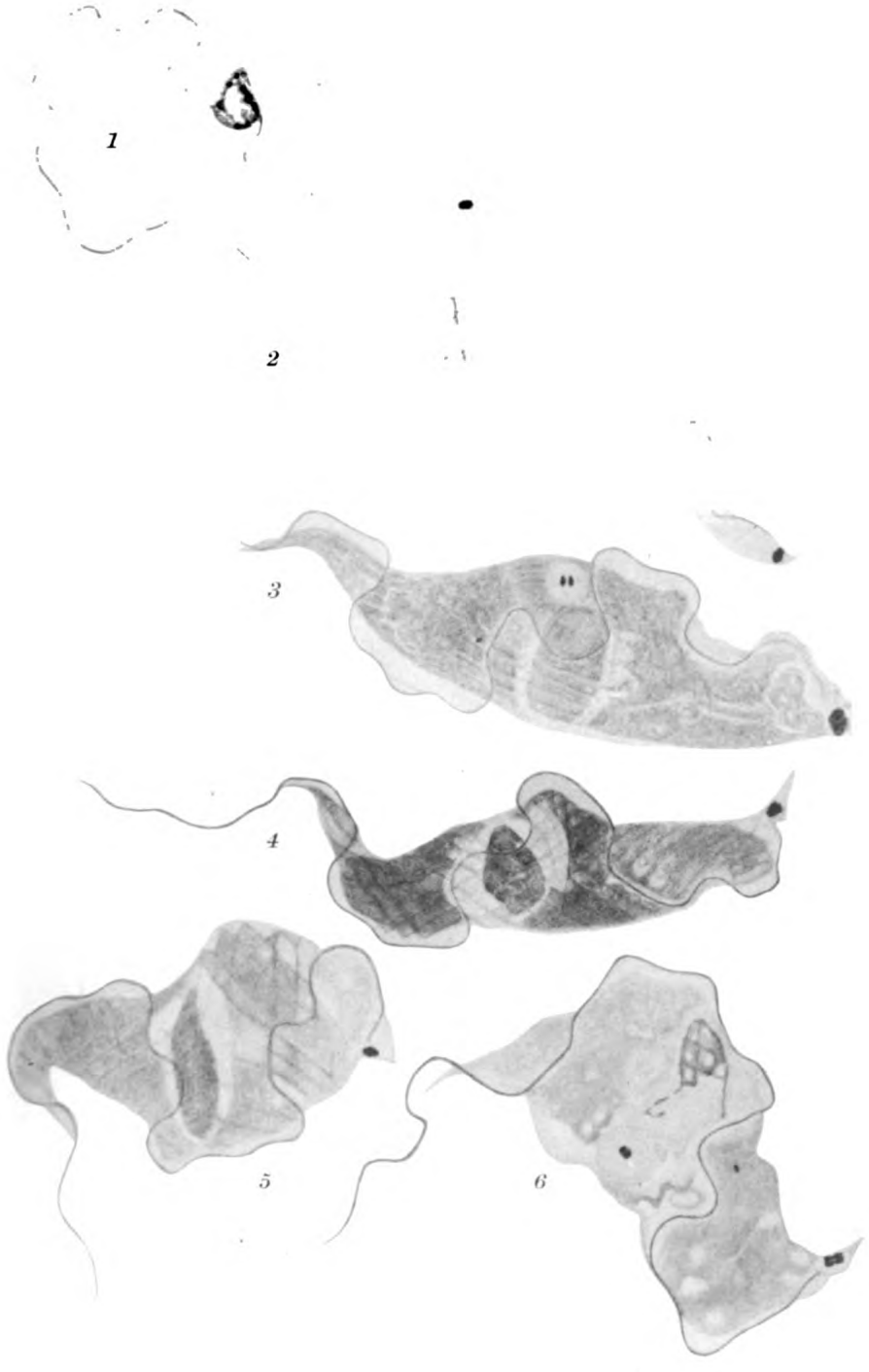
Fig. 2.—" Slender " form.

Fig. 3.—" Broad " form.

Fig. 4.—" Broad " form.

Fig. 5.—" Stumpy " form.

Fig. 6.—" Stumpy " form; evidently degenerated but reproduced to show structure of blepharoplast and nuclear granules.



Dutton, Todd and Tobey.

PLATE XXIV

Trypanosomes.

PLATE XXV*

Fig. 1.—*T. loricatum.* This is apparently an ordinary division form with two blepharoplasts, two nuclei and two flagella. The nuclei are long, sharply-defined sacs, with many small chromatin granules in them.

Fig. 2.—*T. loricatum.* This form has a long nucleus with chromatin massed near both ends. There are longitudinal pleats and a short free undulating membrane.

Fig. 3.—*T. loricatum.* This form has a round nucleus. The longitudinal pleats are present.

Fig. 4.—*T. loricatum.* This form has deep longitudinal pleats. The nucleus is not so long as in figs. 1 and 2, but has the characteristic shape and curves towards the undulating membrane.

Fig. 5.—*T. loricatum.* Lanceolate form. The nucleus is a long and broad band. The undulating membrane runs as a white band in folds down the middle of the parasite.

Fig. 6.—*T. loricatum.* Long "hyla form" (page 324). The body has longitudinal folds. The nucleus is long, and wide near the middle, tapering to both extremities.

Fig. 7.—*T. loricatum.* Long "hyla form." The folds are unfolding. No nucleus is visible.

Fig. 8.—*T. loricatum.* Parasite has become rounded and is about to lose its flagellum.

Fig. 45.—Trypanosome of the tortoise, showing round unstained areas.

Fig. 46.—Drepanidia of the tortoise. The nucleus stains purple and both it and the cytoplasm are denser than in fig. 48.

Fig. 47.—Drepanidium of the tortoise. The nucleus is red and the body has many coarse granules in it.

Fig. 48.—Drepanidium of the tortoise. The nucleus is red and loosely woven, and the cytoplasm is also loosely woven.

Fig. 49. Drepanidium of the crocodile.

Fig. 50. Drepanidium of the snake—common form.

Fig. 51.—Drepanidium of the snake. The nucleus and cytoplasm are loosely woven, and vacuoles are present at both ends.

Fig. 52.—Drepanidium of the snake, showing elongation of the body of the host-cell.

Fig. 53. A curious appearance in a snake's blood (page 304); free form, red dot in the middle.

Fig. 54.—The same in a cell with no drepanidium.

Fig. 55.—The unidentified parasite of the snake—round form.

* Numbers 1-8 are drawn one-half the usual size.

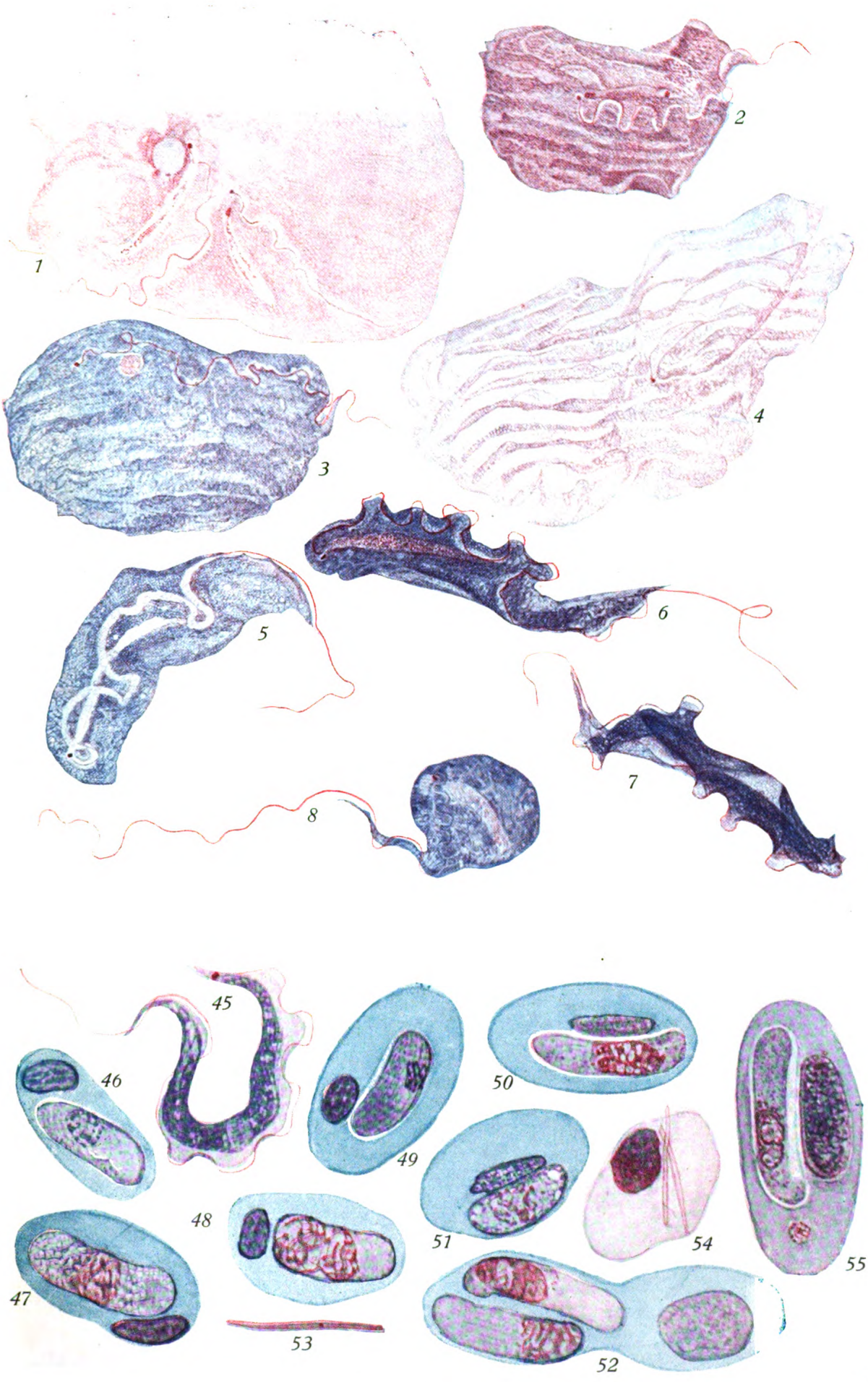


PLATE XXV

PLATE XXVI

Fig. 9.—*T. loricatum.* Wide "hyla form," with folds unfolding. Nucleus indicated by a bent band.

Fig. 10.—*T. loricatum.* Short "hyla form." The nucleus is long and has chromatin massed at both ends. There is an unfolding of the edge of the body at intervals on one side of the parasite.

Fig. 11.—*T. loricatum.* Round form with short curved, narrow white band running from blepharoplast towards periphery. The nucleus has differentiated areas on both sides.

Fig. 12.—*T. loricatum.* Round form, dividing form. Two nuclei, and two blepharoplasts are present.

Fig. 13.—*T. loricatum.* Round form as in fig. 11, but with no differentiated areas in nucleus.

Fig. 14.—*T. loricatum.* Rounded form, about to divide, the flagellum having been lost.

Fig. 15.—*T. loricatum.* Dividing round form; two nuclei, two blepharoplasts, but the cytoplasm not quite divided.

Fig. 16.—*T. loricatum.* Small round division form.

Fig. 17.—*T. loricatum.* A still smaller form than the above.

Fig. 18.—*T. loricatum.* The parasite has divided into a group of 16.

Fig. 19.—*T. loricatum.* Rounded form with division of nuclei and blepharoplasts into four, but with no division of the cytoplasm.

Fig. 20.—*T. loricatum.* Small divisional form that has just acquired a flagellum.

Fig. 21.—*T. loricatum.* Herpetomonas-like form. Note the position of blepharoplast, nucleus, posterior granule, and flagellum.

Fig. 22.—*T. loricatum.* Trichomonas-like form. The anterior extremity is enlarged and round and from it go four flagella.

Fig. 23.—*T. loricatum.* Herpetomonas-like form with large anterior end.

Fig. 24.—*T. loricatum.* This is perhaps a variety of *T. **inopinatum***.

Fig. 25.—*T. loricatum.* This is a common *inopinatum*-like form.

Fig. 26.—*T. loricatum.* An *inopinatum*-like form, wide at the level of the nucleus.

Fig. 27.—*T. loricatum.* A *sanguinis*-like form. Note the wide membrane, the position of the nucleus and the blepharoplast, and the shape of the posterior extremity.

Fig. 28.—*T. loricatum.* A "leaf-like form" (page 322). Note the position of nucleus and blepharoplast, and the shape of posterior and anterior portion of the body.

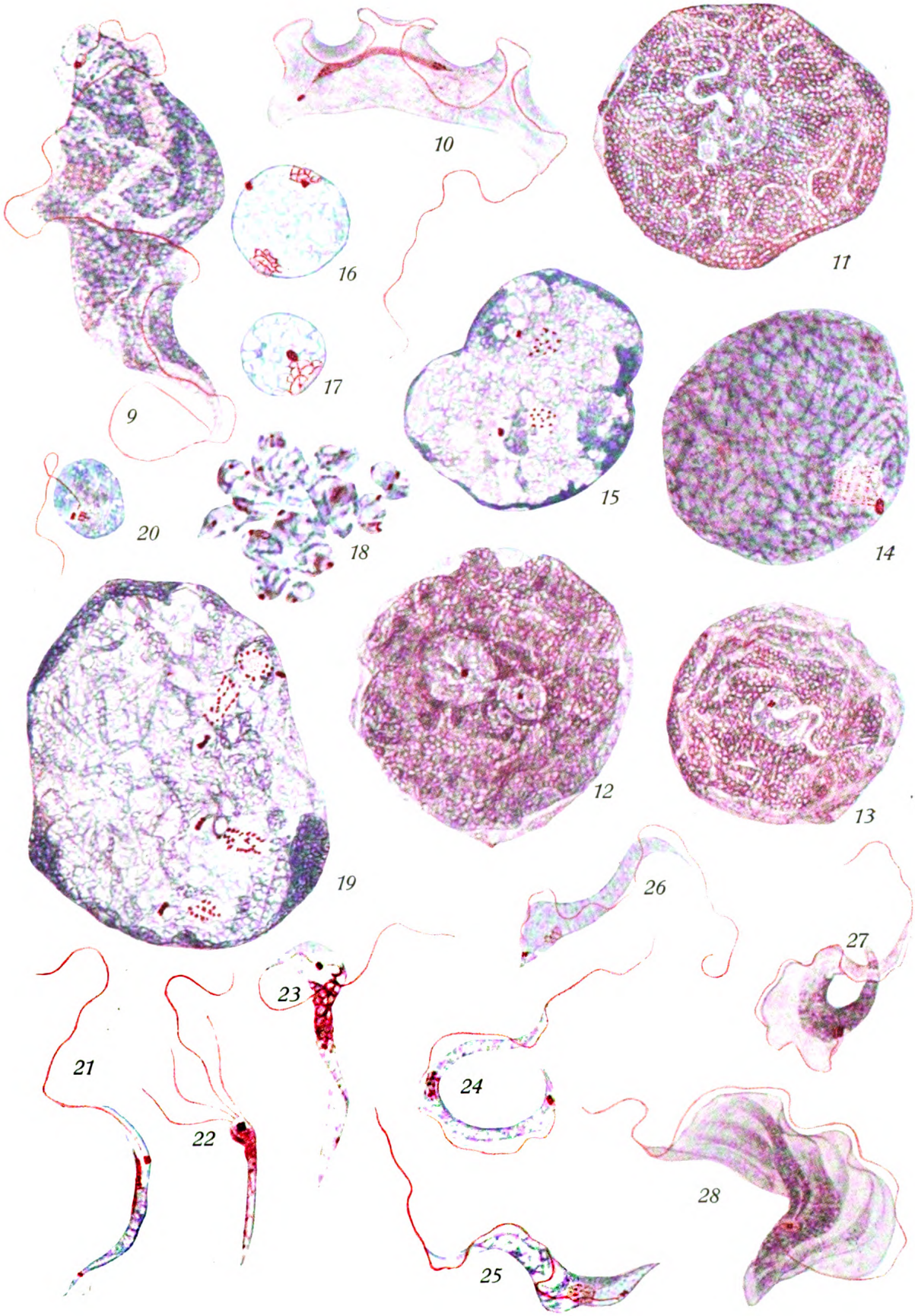


PLATE XXVI.

PLATE XXVII

Fig. 29.—*T. loricatum.* "Leaf-like form" (page 322), with the posterior end arranged like a bud about to open. Note position of nucleus and blepharoplast.

Fig. 30.—*T. loricatum.* "Leaf-like form" unfolding. Note position of nucleus and blepharoplast, and shape of anterior and posterior portions of body.

Fig. 31.—*T. loricatum.* The "leaf-like form" has completely unfolded into an adult type of *T. loricatum.*

Fig. 32.—Intermediate form between the "leaf-like form" of *T. loricatum* and *T. mega.*

Fig. 33.—*T. mega.* A form with red staining granules in the cytoplasm. The nucleus is faintly stained.

Fig. 34.—*T. mega.* Same form as fig. 33, but with more red granules in the cytoplasm.

Fig. 35.—*T. mega.* The posterior part of the body is irregularly contracted. Note the shape of the nucleus.

Fig. 36.—*T. mega.* A somewhat generally contracted form. Note the shape of the nucleus.

Fig. 37.—*T. mega.* A very much generally contracted form. Note the shape of the nucleus.

Fig. 38.—*T. mega.* A variety of *T. mega* with coarse reticular structure.

Fig. 39.—*T. mega.* A much elongated form. Note the character of the nucleus, and the difference between the part of the body anterior to the nucleus and that posterior to the nucleus.

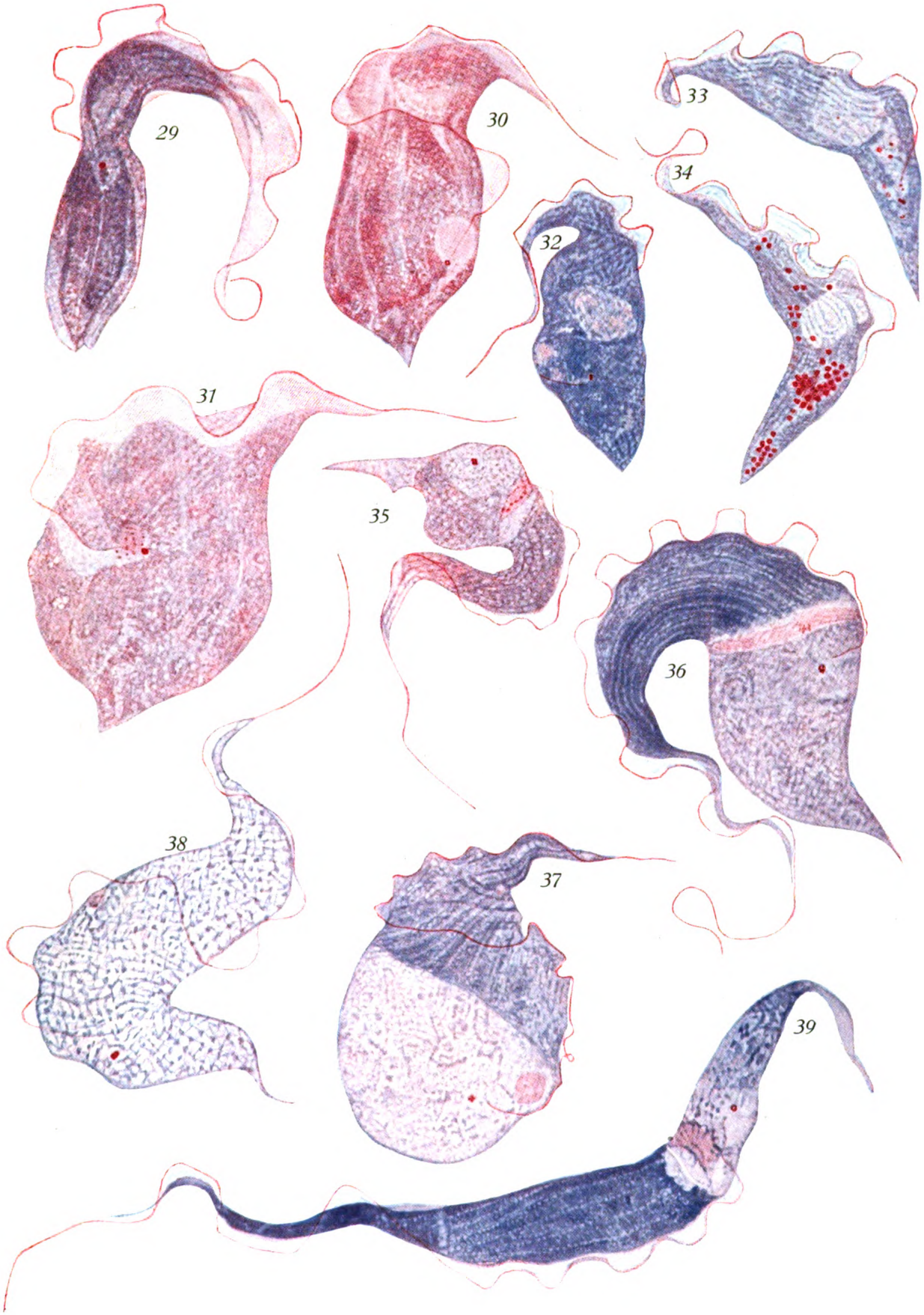

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PLATE XXVIII

Fig. 40.—Small form of *T. karyozeukton*, showing blue spiral line running from blepharoplast to posterior extremity, and the thick wide undulating membrane.

Fig. 41.—Medium form of *T. karyozeukton*, showing granule at posterior end of thickened edge of undulating membrane.

Fig. 42.—Large form of *T. karyozeukton*.

Fig. 43.—*T. karyozeukton* coiled, with the anterior end free.

Fig. 44.—Tighter coil of *T. karyozeukton*.

Fig. 56.—A curious appearance in a snake's blood (page 304) in a cell with a drepanidium.

Fig. 57.—The same lying upon a degenerated drepanidium.

Fig. 58.—The same with short rods of unequal length.

Fig. 59.—Intracellular drepanidium, showing excretion at both extremities within capsule.

Fig. 60.—Encysted drepanidium, found free in serum, showing excretion at the extremities.

Fig. 61.—Division form of drepanidium. The cytoplasm has divided synchronously with the chromatin.

Fig. 62.—Division form of drepanidium, dividing into two.

Fig. 63.—Division form of drepanidium, with the divisions almost separated.

Fig. 64.—Division form of drepanidium, five divisions.

Fig. 65.—Division form of drepanidium with four divisions.

Fig. 66.—Division form of drepanidium on the way out of the erythrocyte.

Fig. 67.—Common form of drepanidium, almost out of an erythrocyte, showing the eaten-out character of the host cell.

Fig. 68.—Common form of drepanidium, showing division of nucleus into two, connected by a red line.

Fig. 69.—Common form of drepanidium with dividing nucleus.

Fig. 70.—Common form of drepanidium with constriction at one end of the body, showing one method of movement.

Fig. 71.—Common form of drepanidium with two constrictions.

Fig. 72.—Young parasites of the small form of drepanidium.

Fig. 73.—Small form of drepanidium, showing nucleus and the blue spiral line running from a chromatin granule of nucleus to anterior extremity of body, × 4,000 to show structure in detail.

Fig. 74.—Small form of drepanidium, showing chromatin granules at posterior end, also a few anterior to nucleus, × 2,000.

Fig. 75.—Free division form of drepanidium.

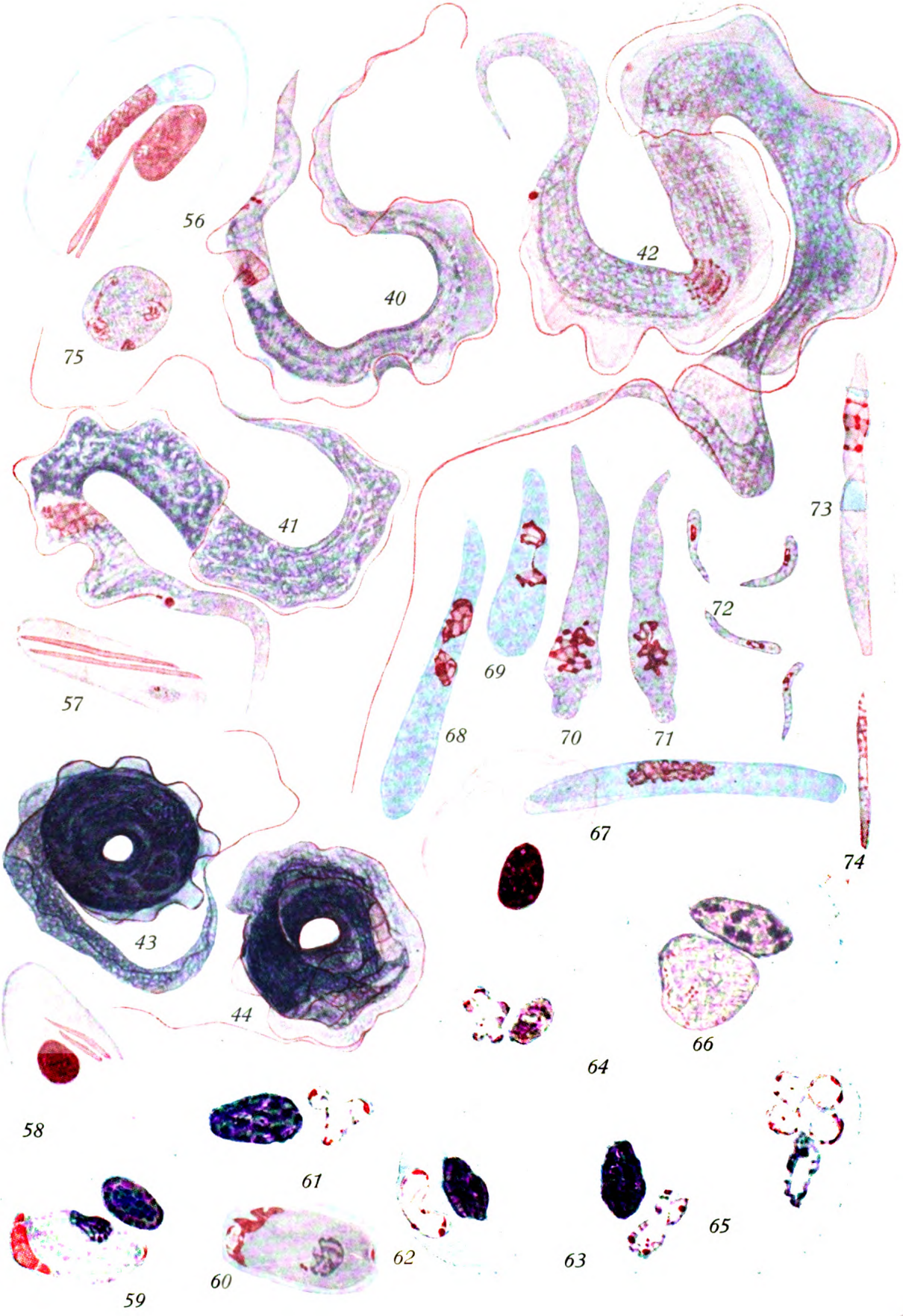


PLATE XXVIII.

PLATE XXIX

Fig. 76.—Division form with one larger mass of chromatin and several chromatin dots around it.
Fig. 77.—Division form with four masses of chromatin.
Fig. 78.—Division form with sixteen masses of chromatin.
Fig. 79.—Division form with twelve masses of chromatin and the division into well-defined "spores."
Fig. 80.—Double infection with rosette division forms.
Fig. 81.—Common form of drepanidium with division form beside it in same host-cell.
Fig. 82.—Common form of drepanidium with division form at an earlier stage of development.
Fig. 83.—Young form of large drepanidium.
Fig. 84.—Drepanidium showing one globule situated near the nucleus.
Fig. 85.—Drepanidium with about 100 refractile globules in it.
Fig. 86.—Large form of drepanidium.
Fig. 87.—Large form of drepanidium, showing body folded upon itself.
Fig. 88.—Large form of drepanidium, only partly folded upon itself.
Fig. 89.—Large form of drepanidium with 34 refractile globules.
Fig. 90.—Large form of drepanidium with two globules situated near nucleus.
Fig. 91.—Drepanidium with 11 globules situated near the nucleus.
Fig. 92.—Drepanidium with large refractile globules.
Fig. 93.—An unidentified parasite of frog with crystal.
Fig. 94.—An unidentified parasite of frog, showing a blue area sharply defined.
Fig. 95.—An unidentified parasite of frog, with crystal in nucleus.
Fig. 96.— „ „ with well-defined blue area.
Fig. 97.— „ „ apparently free in the serum.
Fig. 98.— „ „ with three crystals.
Fig. 99.— „ „ triple infection.
Fig. 100.—Cytomoeba free in serum.
Fig. 101.—Cytomoeba in cell.
Fig. 102.— „ „
Fig. 103.— „ „
Fig. 104.— „ „
Fig. 105.— „ „
Fig. 106.— „ „ round form.
Fig. 107.— „ „
Fig. 108.— „ „ round and long forms.
Fig. 109.— „ „ long form.
Fig. 110.— „ „ round forms breaking up nucleus.
Fig. 111.— „ „ round form breaking up nucleus.
Fig. 112.— „ „ long form.
Fig. 113.— „ „ long form.
Fig. 114.— „ „ round form.
Fig. 115.— „ „ long form.

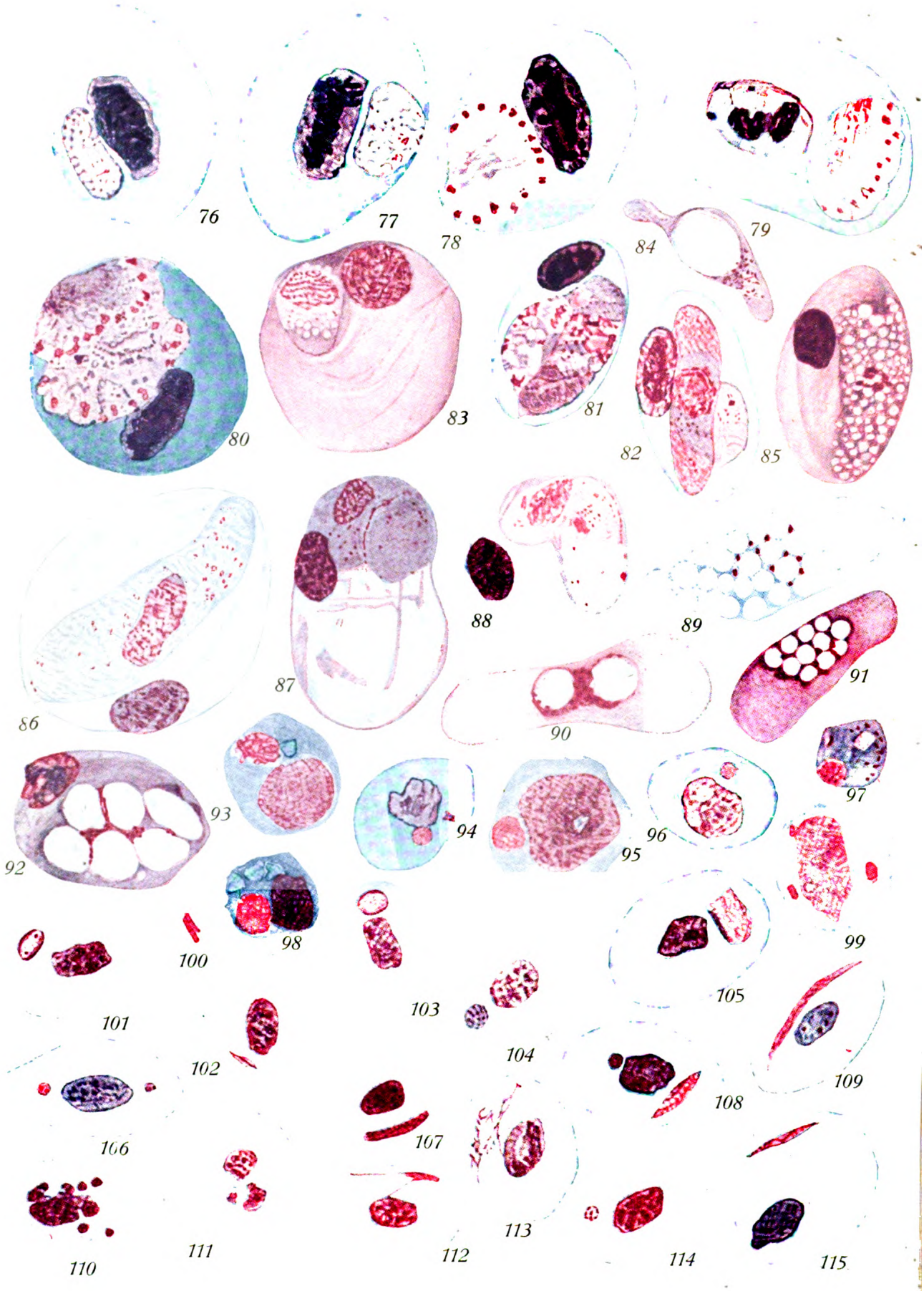

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PLATE XXIX.

PLATE XXX

Fig. 1. Small form, showing arrangement of the chromatin granules of the nucleus and of the granules of the blepharoplast.

Fig. 2. Small form, showing early stage of division. The chromatin granules of the nucleus have gathered into two masses, and the granules of the blepharoplast have changed their position.

Fig. 3. Medium form, showing arrangement of the chromatin granules of the nucleus and of the granules of the blepharoplast.

Fig. 4. Medium form, showing striations.

Fig. 5. Long form, showing arrangement of the chromatin granules and karyosome of the nucleus, the longitudinal striations, the granules of the blepharoplast and the vacuoles anterior to it.



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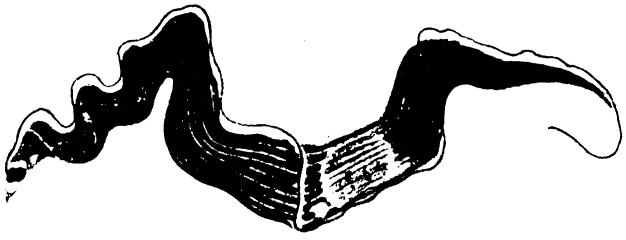
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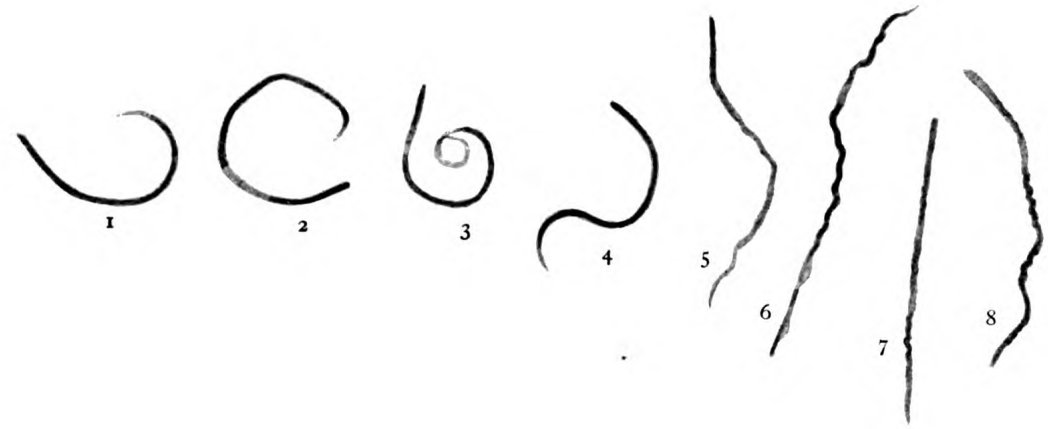
Dutton, Todd, Tobey

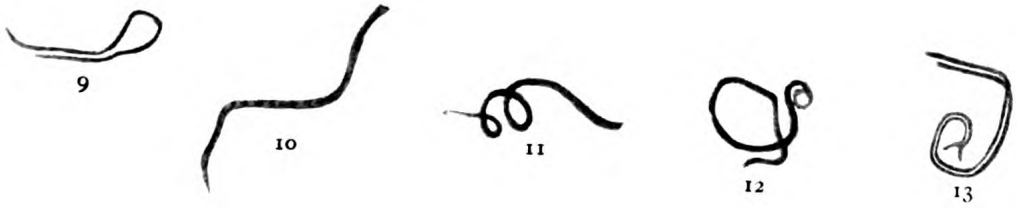
PLATE XXX

Hemoflagellate

PLATE XXXI

Spirochaeta jonesii from *Clarias angolensis*, see page 338.







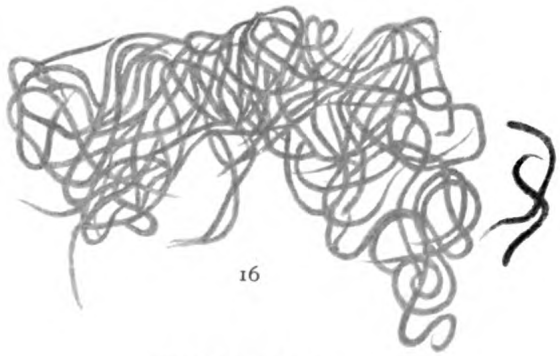


PLATE XXXI

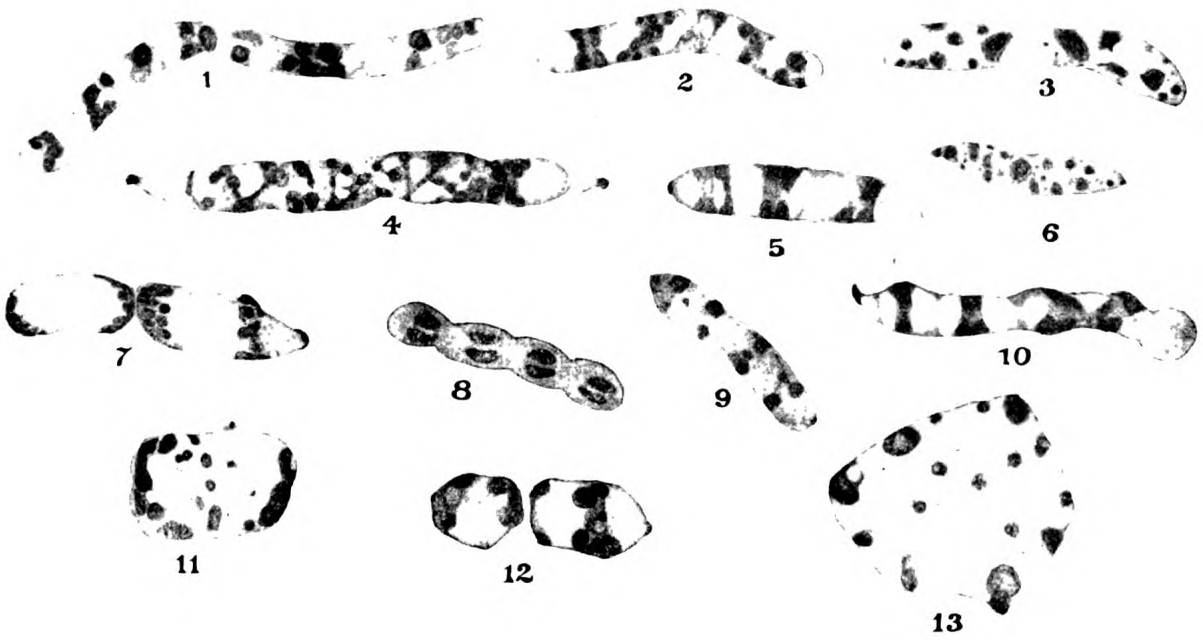
Dutton, Todd, Tobey

Hemoflagellate

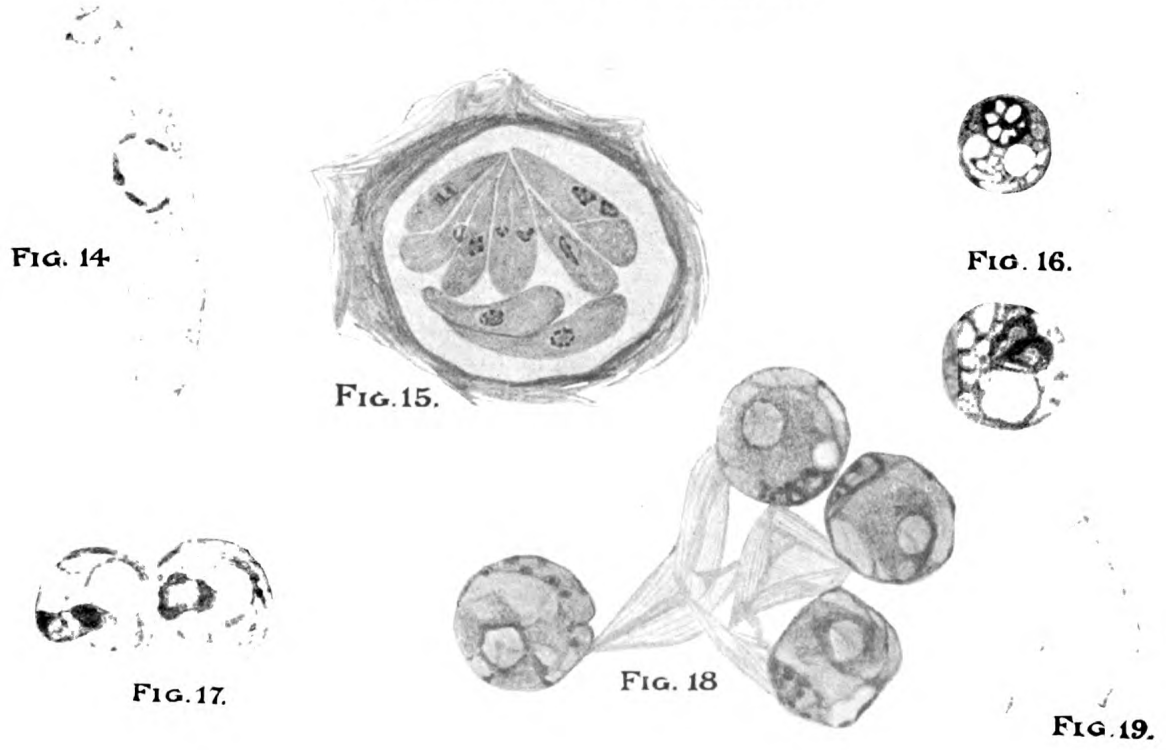
PLATE XXXII

Myxosporidium heibergi from *Glossina palpalis*, see page 342.

Coccidium ——? from *Pyretophorus costalis*, see page 339.



MYXOSPORIDIUM HEIBERGI.



COCCIDIUM OF PYRETOPHORUS COSTALIS.

YAWS

YAWS

BY

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(*Received May 13th, 1907*)

THE PREVALENCE OF SYPHILIS IN THE WEST INDIES

The most striking feature of the medical practice in the West Indies is the prevalence of tertiary syphilis and infantile inherited syphilis among the labouring class. In some of the islands this prevalence is quite extraordinary.

On the contrary, in the classification of diseases returned by the Colonial medical officers, as well as in the reports of hospitals, syphilis is credited with so few cases that it is incredible that the figures can be correct. In part this is due to the custom of classing diseases by the local manifestations, e.g. iritis, necrosis, paralysis, &c. The cases are thus scattered through the classification, and even if their true nature has been recognized it is not shown. But it is nevertheless a fact that great misconception does exist in the minds of many of the medical profession in the West Indies, and no doubt elsewhere in the tropics, as to the extent to which syphilis is responsible for the sicknesses that occur. The chief causes of this failure to recognize the disease are ignorance of the original nature of syphilis and the tendency to regard it as a venereal disease.

Failing to attribute a given lesion to syphilis the medical man naturally turns to tubercle, the effects of which on the natives of the tropics are in consequence vastly over-rated. Tubercular bone and joint disease is rare among negroes. Lupus I have never seen, though I have seen cases classed as such, the scars and subsequent history of which abundantly proved the syphilitic origin of the lupoid ulceration. Phthisis is very common among the negroes in some West Indian Colonies, but in view of the undoubted rarity of other

tubercular troubles, I am inclined to believe that a great deal of the phthisis is really syphilitic. I have several times been agreeably surprised by the recovery of a phthisis case. Without actually diagnosing syphilis, one gets such a belief in iodide, and uses it for such a wide range of complaints, that some of the lung cases have had the good fortune to be accidentally cured in this way. It is a safe thing to treat all supposed phthisis with iodide if the tubercle bacillus cannot be found or if there is no opportunity to search for it. But even after discounting for pure syphilis of the lung, there remains a number of tubercular cases quite out of proportion to the amount of tubercle of other tissues.

With respect to the West Indies I can personally vouch for the prevalence of syphilis in the islands St. Croix (Danish West Indies), St. Kitts, Nevis, Antigua, St. Lucia and St. Vincent. In Barbados I think there is not so much of grave syphilis, but the disease is widely spread nevertheless. Dr. W. J. Branch, after 25 years' practice in St. Kitts, said in reply to a query, "nearly every black or "coloured person on St. Kitts has, or has had, syphilis in some shape "or other, congenital, acquired or both."[1] I had five years' experience in district and hospital work in that island, and can assure the reader that though this estimate is true, the condition of the population of St. Vincent is worse. Here I have seen a man in middle life with tertiary scars dating from childhood, a scar of an old penile chancre, and a new eruption of secondaries. That is to say, in St. Vincent people may be "thrice dipped" in syphilis. Second infections are the rule of life if an individual comes to mature years. The combination of secondaries and tertiaries is very commonly seen.

In my annual hospital report for 1902-03 I wrote:—

"When to this moral state is added a profound and universal "saturation with syphilis and depletion by ankylostoma, it may easily "be understood that the present labouring population of St. Vincent "is as diseased and pauperized as any in the world. The effects of "syphilis depend on the nutrition of the patient; so that where there "is a soil as suited for the exuberant manifestation of the disease as "obtains here, it is not surprising to note the disablement and "increasing pauperism due to syphilitic ulceration and necrosis, "parasyphilitic paralysis, and degenerative neurosis.

1. Nicholls' Report, page 167.

"Medical men from abroad, visiting St. Vincent, are struck by "the prevalence of disfigured and noseless faces, and the pauper "asylum is a museum of remnants left by syphilitic disease and the "surgeon's knife."[1] This is a lurid picture, but it is paralleled by the condition of more than one other West Indian colony.

Dr. Blanc, of Tobago, bears witness to the extensive spread of syphilis in that island. "In connection with this question, I may say "that in Charlotteville and Speyside, where yaws was so prevalent, "a very large proportion of the population suffer from syphilis."[2]

Dr. S. Branch, in his hospital report from St. Lucia for 1904-05, speaks as strongly of the prevalence of syphilis there as I do of St. Vincent.

These, after all, are only statements of opinion, but though figures are proverbially deceptive, I am able to adduce some statistics in support of my assertions with respect to St. Vincent. We may take these as practically representative of the state of affairs in the West Indies generally; for with few exceptions they all belong to one type; the poverty, the race of the masses, the climate, and the geologic structure are the same.

It was agreed by the Medical Officers of St. Vincent that all cases of tubercle and syphilis were to be classed as such in the monthly returns of cases treated, and not scattered under the different organic systems as local diseases. This has been done since July, 1905, and I can show some reliable figures for twelve months' district work in the Colony. The population of St. Vincent is estimated at 45,000. The hospital has a daily average of 55 to 60 patients. In the four years I have been in charge of the hospital there have been 3,269 admissions for all causes. This number is rather swollen by the cases of ankylostomiasis, many of which are admitted twice or several times for two days in order to take thymol. In these four years there have been 630 admissions for syphilis, of which 42 were for primary. Only about 20 of these cases were suffering from yaws, so that I have not materially swelled the total by including this condition with syphilis. The syphilis cases therefore form about 19 per cent. of the total admissions. When it is remembered that one naturally avoids filling one's hospital beds with chronic cases, such as those

1. Hospital Report, St. Vincent. Colonial Reprints, No. 20.
2. Nicholls' Report, p. 166.

suffering from tertiary ulceration, and that ankylostomiasis has run the total to an abnormally high figure, it must be admitted that this percentage shows well the prevalence of syphilis.

For some time I conducted an out-patient department at the hospital, as a single-handed effort to deal with some of the tertiary cases of the island. Between July, 1903, and March, 1905, that is, in twenty-one months I treated 600 cases of syphilis.

In the four years of hospital work referred to, a large number of amputations have been necessitated by syphilis. Of 51 amputations of the leg at the seat of election, 27 were performed for syphilis and 11 for elephantiasis. And even of the latter several were brought to the knife by extensive syphilitic ulceration.

In the reports of the Medical Officers for the district work for twelve months—July, 1905, to June, 1906—no less than 1,996 cases of syphilis are returned. This for a population of 45,000 is equal to 4·4 per cent. per annum of the inhabitants. Of these I attended myself 539 cases, which form about 21 per cent. of all the district cases treated by me in that time. It must be understood that these figures referring to syphilis have not been swelled by the inclusion of the cases of yaws. Though it is true that only one Medical Officer in St. Vincent now returns yaws as such, yet this condition is not often brought to our notice, and the few cases of frambesial eruption which we have treated would not affect the general truth of the figures. The one Medical Officer referred to whose district is that most infected with yaws, only returned 24 cases in the twelve months.

There is no compulsory medical certification of death in St. Vincent, except for infants under one year, but nearly all the deaths are certified by medical men. I went through the register for two years—March, 1904, to March, 1906—to pick out and classify the causes of death. From the vital statistics I may quote the following:—

Total deaths	1,418
Still Births	233
Syphilis	56
Hereditary Syphilis (infants)	149

Syphilis, therefore, causes on the average about 14 per cent. of the deaths in St. Vincent.

A high infant mortality (25 per cent. of the total deaths in St. Vincent) and a large number of still-births are observed all over the West Indies. These alone are evidence of the prevalence of syphilis, though there are other causes—illegitimacy, ignorance and poverty—which contribute to raise these rates.

Hereditary syphilis in infants is a matter which presses hardly on Government Medical Officers who are required to attend the children of labourers free, as they do in many West Indian Islands. In the twelve months' period referred to above, I saw 110 infants suffering from inherited disease out of a population of about 5,000. In the whole Colony in the same period there were returned 512 cases of hereditary syphilis. This is in a higher ratio to the total cases of syphilis than in my practice. I attribute this difference to my own tendency to class syphilis of children and young persons as acquired, only counting syphilis in infants as certainly or probably inherited. Comparing the ratios otherwise, I find a striking agreement in the figures relating to syphilis returned by myself and the other Medical Officers.

To recapitulate the proofs of the prevalence of syphilis in St. Vincent, we note

1. 19 per cent. of the admissions to hospital are for syphilis.
2. 4·4 per cent. of the inhabitants are attended yearly for syphilis by the district Medical Officers.
3. In a small hospital in a small colony no less than 27 legs were removed on account of syphilis in four years.
4. The cases of syphilis form 21 per cent. of the district cases seen by one Medical Officer.
5. Syphilis is responsible for 14 per cent. of the deaths in the Colony.

I take St. Vincent as a representative Colony in which the disease is well developed, though probably not much more so than in some other West Indian Colonies.

If now we examine the stages of syphilis seen by the medical men in St. Vincent, we shall find a preponderance of the tertiary and what seems to be an inadequate proportion of the primary stage. The figures for the same twelve months' period are as follows:—

Primary	35
Secondary	373
Tertiary	1,076
Hereditary	512

I may admit that the primaries include several (say four) non-venereal cases returned by myself.

It is true that the primary lesion, even when venereal, is frequently a very trivial-looking thing to the patient, but many such sores are aggravated by the dirty habits and incontinence of the black patients, and must come to hospital for surgical treatment. In St. Kitts we operated very often on such cases. There are, therefore, probably not many chancres of the genitals, in St. Vincent at any rate, which are not brought to the surgeon's notice.

This paucity of venereal chancres in the tropics is referred to by Numa Rat, whose experience extended to the West Coast of Africa as well as the West Indies. "And I may add that it is very rarely "indeed that I have seen the initial lesion of syphilis in a black man "in any country. Either the syphilitic chancre in the negro is a much "less serious affair than it is in a European, or the former considers "it too trifling, whatever its severity, to trouble the doctor about. It "is reasonable to assume that in a place like St. Kitts, in which "syphilis prevails so generally, the initial lesion of the affection would "be modified by the presence of an inherited syphilitic taint."[1]

It may justly be contended that 31 venereal primaries in 1,996 cases of syphilis is an inadequate proportion. And, again, in four years only 42 cases of primary have been admitted to hospital. It seems certain that the venereal chancres do not account for the amount of syphilis in the Colony, and it will be my endeavour to suggest an explanation in the following pages, which I believe applies equally to all tropical countries.

TERTIARY SYPHILIS IN EARLY LIFE

While we were speaking of tertiary syphilis, the reader no doubt has had in his mind the syphilis of adults. But we have now to consider another phase of the question. A great deal, if not most, of the tertiary in the West Indies is seen or has begun early in the life of the patients, and much of the juvenile syphilis is of a very severe character.

The large infant mortality indicates what has become of most of

1. Report on Anguilla. Colonial Reprints, No. 19.

the inherited syphilis, and it is well known that nowhere is the occurrence of inherited taint in proportion to the number of infected parents. If this were not so the human race would have disappeared at an early period of its existence. I have rarely seen a black or coloured child over three years of age whose syphilis was undoubtedly inherited. Eye lesions, sabre shins and cranial malformations occur, but by no means commonly among the juvenile cases of syphilis. Hutchinson's teeth I have never seen in a dark-skinned person, and doubt if this mark affects the negro race.

Moreover, the manifestations of disease in young persons are usually of a severity that one does not associate with inherited taint. Under the unfavourable conditions of child life among the black peasants, the bad cases of inherited disease die early. The milder cases we should not expect to run to such virulent tertiaries as we commonly see. Those writers who try to prove that some of these lesions, such as necrosis of the palate, are not caused by syphilis urge the youth of many of the patients, and thus they indirectly admit the improbability of their being due to inherited taint. I can therefore produce these writers in evidence. J. Numa Rat is perhaps the most careful and accurate observer of yaws. He does not believe that this condition has any relation to syphilis, but finding a prevalence of lesions apparently identical with those of tertiary syphilis, he did not attribute them to inheritance, but to yaws itself.

The very common lupoid ulceration is rarely ascribed to syphilis in the West Indies for the same reason of the youth of the patients; it is called lupus. Yet it invariably gets better under iodide, and often spontaneously. Just as common in St. Vincent is ulceration and necrosis in the throat and nose, affecting young people more frequently than adults. In a recent correspondence in the Journal of Tropical Medicine, James Leys[1] and Numa Rat[2] discuss this condition, which the former found as prevalent at Guam in the Philippines as it is in the West Indies. It is said to be very common also in Fiji. Leys suggests the name "rhinopharyngitis mutilans" for the condition, and both the above writers agree that it cannot be syphilis because the patients are often young, fourteen to nineteen years. As a matter of fact, sexual life begins so early with the dark races that

1. Jour. Trop. Med. February 15th, 1906.
2. Jour. Trop. Med. May 1st, 1906.

a patient of sixteen years could easily have had time to reach this stage of tertiary from a venereal infection. And in my own experience in St. Vincent most of the few females I have seen with venereal primary were about or under this age. But I have seen rhinopharyngitis in patients as young as nine or ten years. Both these writers ignore the possibility of non-venereal infection in childhood, and neither even raise the question of inheritance.

The rhinopharyngitis as described by Leys and accepted by Rat is identical with that we have in St. Vincent. It is such a characteristic tertiary that it would be diagnosed as such without a moment's hesitation if seen in a single individual. But it is very common in some places, e.g. certain parts of the West Indies, Guam, Fiji, while in other places where there is just as much syphilis it is only occasionally seen. It is this peculiarity of distribution which makes the difficulty. I have pointed out that the damp, hot, wooded ravines of volcanic islands in the tropics are infested alike by yaws and rhinopharyngitis. The same climatic conditions determine the occurrence or favour the development of both. The point to which attention has not been sufficiently directed is that syphilis is modified by circumstances of climate, race, personal habits and constitution, &c., so as to manifest itself in various forms. In St. Vincent, for example, lupoid destruction of the face is the rule on the Windward side of the island, and necrosis of the palate and turbinated bones on the Leeward. It is incorrect to judge of syphilis of dark races in a tropical hothouse by the standard of syphilis in Europe. So certain is it that rhinopharyngitis mutilans is a tertiary that its presence in Fiji should be conclusive that the supposed freedom of that group from syphilis was a delusion, if such proof was ever seriously required.

The condition is this; there is ulceration of tonsils, fauces, pharynx, palate or nose, progressing to destruction and accompanied usually by necrosis of the hard palate, the turbinated bones and vomer. Any or all of these parts may be involved. The nose at length may fall in, but the ulceration does not usually reach the skin of the face. One may see the buccal, nasal, and pharyngeal cavities thrown into one space, lying between the tongue and the base of the skull, and lined by a vast green-gray ulcer relieved by blackened remnants of necrosed bone. The condition does not appear to kill readily, for the

1. Jour. Trop. Med. May 15th, 1906.

patients continue slowly losing tissue for years when untreated, and it may heal even after most extensive destruction has taken place. There is just as frequent syphilitic ulceration, but of the skin, in other places where the rhinopharyngitis is not so common.

Hutchinson describes the same lesion as being not infrequent in the young, and attributes it to inherited taint. It is remarkable, however, that he does not usually find the other signs of heredity, notched teeth and keratitis, in such cases. In view of the facts which I shall later on present, I have taken the liberty of disagreeing with the master's opinion, and believe that this severe tertiary is usually the result of acquired syphilis.

According to Hutchinson, the tertiaries of inherited syphilis commonly seen are interstitial keratitis at about the age of puberty; deafness; periosteal nodes between the ages of, say, eight to ten years, sometimes ending in necrosis; lupoid ulceration and ulceration of the pharynx and palate. I have only seen three instances of deafness in juvenile syphilitics in St. Vincent. Interstitial keratitis is rare; I can only remember one case. Perhaps the negro race is not prone to it, though iritis and other eye affections are sometimes seen in acquired syphilis of blacks. Periostitis and synovitis in young persons and children are common enough, but these belong as much to the acquired disease. Lupoid ulcerations and ulceration of the nose and throat are, as we said before, very common in young persons. These also occur in acquired syphilis.

But it is not, I believe, recognised that cutaneous gumma and subcutaneous gumma leading to gummatous ulcers occur or are at all common in the inherited disease. So frequent is the former in the West Indies that it is well known to the natives under the names "blind-boil" and "blue-blister." It is just as common in children as in adults.

The terrible and long-lasting ulcers often met with in the young peasants are sometimes superficial and lupoid or serpiginous, but more commonly they are deep subcutaneous gummata. One may see the early stage when there is a fluctuating tumour so likely to be mistaken for abscess. I have myself cut into such a gumma in a child of eight years, under the impression that it was a cold abscess, and found the characteristic soft pale tissue exuding a gummy secretion from the cut surface. The frequency of these tertiaries is, I think,

conclusive of the occurrence of youthful acquired syphilis, for if even possibly hereditary, they can only be rarely so.

On the whole, therefore, while there is a great deal of congenital syphilis of infants, yet I am not satisfied that the amount of youthful tertiary which we see can be accounted for by inheritance. As before remarked, most of the congenital disease disappears by the death of the infants. What is left is presumably of a milder type, and would not produce such virulent and lasting tertiaries. When it can be shown, though this is anticipating my argument, that innocent syphilis is of frequent occurrence (apart from yaws), I feel I am justified in saying that the juvenile tertiary as seen is not the result of inherited taint.

I have for some time been in the habit of jotting down short notes of syphilitic cases, though this has not been kept up regularly, and there are long and frequent hiatuses in my record. Yet out of the notes, such as they are, I can place before my readers enough cases to illustrate the several points to which I wish to call attention.

I think it will be agreed that the cases following here read much more like examples of acquired than hereditary syphilis in spite of the youthfulness of the patients. I have been limited by selecting cases in which there was no history of yaws.

CASE I.—B. I.; 20 years; Black. No note of earlier history.

June 6th, 1903. Pigmented scars on both legs. Deep adherent scar over the lower end of the left fibula. Has had an ulcer of the right leg for some years. At present this is an enormous sloughing ulcer involving half the circumference of the leg, which is much enlarged. The rest of the circumference of the leg is cicatrized. Refused amputation. This is clearly a case of tertiary of many years' standing in acquired disease. Yet at the time when she must have had the primary she was rather too young for venereal infection.

CASE 2.—W. S.; 13 years; Hindu.

June 1st, 1906. Deep scars on both knees. On the right ankle an extensive and adherent scar. Said he had this more than four years ago (dated by the volcanic eruption). A large foul tertiary ulcer covers the back of the right foot. This has been of two years' duration. The tertiary here dates from the age of nine years.

CASE 3.—D. S.; 10 years; Black.

July 10th, 1903. Pigmented scars of "blind-boils" on both legs. Several superficial ulcers on the left leg. Swelling of left ankle, and periostitis of lower end of left fibula. Enlarged glands in both groins, those of left side tender. Hard glands on both epicondyles of elbows.

The blind boils are small indolent ulcers about 6 to 10 mm. wide, and circular. They are commonly multiple and symmetrical in arrangement. They begin as a pimple which breaks down and suppurates, leaving a shallow punched

ulcer, which is very resistant to treatment. They seem to belong to the "intermediate" stage of Hutchinson. This case is one of early tertiary in acquired disease, at an age which precludes the possibility of venereal infection.

CASE 4.—A. M.; 12 years; Black. Denies yaws.

July 2nd, 1903. Left tibia sabre-shaped. Right with a very large node. Large scar on back of right thigh. Large ulcer inside the upper lip. Columna nasi has disappeared, and a large ulcer covers the surface of the nasal cavity, eroding the alae nasi. Several scars of previous skin gummata on the inner side of nates. Here it may be admitted there is a probability of heredity indicated by the sabre shin.

CASE 5.—W. C.; 18 years; Black. Has been five years in the Pauper Asylum as a hopelessly disabled person.

October 11th, 1902. Left arm has been amputated below the elbow. Elbow scarred and ulcerated. Left leg is a scar from hip to toe, with the knee and ankle contracted in web-like keloids. Right leg is scarred below the knee. Both feet are masses of ulcers and bone disease.

After more than two years' treatment with iodide, and amputation of the left thigh, he was completely healed and left the Asylum. He has remained well to the present. I think that hereditary syphilis is not known to produce so frightful a condition.

CASE 6.—J. L.; 22 years; Black. Sent to the Pauper Asylum at the age of 16 years.

October 17th, 1902. Both knees and elbows stiff and contracted; extensive scarring and lupoid ulceration on both shins. Multiple nodes on forearms and humeri which give them a distorted appearance. Hard palate perforated. Deaf.

This case had since healed, but lately developed intense headaches and very marked mental dulness. There is no question of venereal infection in one who is already hopelessly disabled at 16 years.

CASE 7.—J. B.; 22 years; Coloured. Was disfigured at the age of 14 years. Was sent to the Pauper Asylum at 17 years.

October 17th, 1902. Nose is gone, lower lids retracted by scarring. Side of upper lips drawn upwards. Face scarred generally. Left leg extensively scarred by gummatous ulcers on the lower half of the shin. Ulceration of throat. Since healed under potassium iodide and sent out. Here again the early age at which the patient was considered incurable precludes the venereal infection.

CASE 8.—W. S.; 9 years; Black.

October 23rd, 1902. Large keloid on right elbow and a scar on the forearm. Large keloid scar on left knee. Scar on outer malleolus. Large gummatous ulcer and scars on right ankle. The scarring here indicates deep or subcutaneous gummata which had been appearing during several years.

CASE 9.—R. S.; 14 years; Hindu.

November 7th, 1902. Extensive scar all round the left elbow and half of the arm. Extensive scar on the back of the hand, and a scar on the wrist. Two fingers contracted and a few small ulcerated spots on the fingers. Extensive scars at the back of the right elbow, back of lower forearm, and back of hand. Large scar over the front of left knee, and another of the calf. Scarred all over the lower end of the tibia. Foot a distorted mass of scars and bone disease, with one large and several small ulcers. Scars on right knee and on lower end of tibia. Large ulcer of ankle. Seven superficial scars on cheek, nose and lip. Septum nasi gone. Was treated in hospital with a view to amputation of the

left leg, but when considered fit he declined operation and was discharged. He has not been heard of since. This is a class of case that is never seen in Europe. I have notes of several as severe cases in St. Vincent.

CASE 10.—L. B.; 9 years; Black.
November 21st, 1902. Perforating ulcer at junction of hard and soft palate. A very young case of "rhinopharyngitis."

CASE 11.—C. A.; 16 years; Black. Says, "never had yaws."
March 1st, 1906. Scar of velum palati with a perforation and a cleft of the palate. Scar on the face, one on the arm, and several on the legs, of tertiary ulcers. Two of these involved loss of bone from the tibia. Gummatous ulcer of one foot and the other leg.

CASE 12.—A. S.; 12 years; Black.
February 11th, 1903. Fistula of lachrymal duct from disease of maxillary bone. Left ala nasi deformed and nostril contracted by a scar. Cartilage of septum has gone. There is an ulcer of soft palate and fauces. Uvula has disappeared. Another young case of "rhinopharyngitis."

CASE 13.—E. W.; 12 years; Black. Had disease of the nose two years ago.
November 22nd, 1902. Ulcer and necrosis of palate. Ulcers of right thigh, knee and skin. Ulcer and bone disease of first metatarsal. Ulcers of left thigh, knee and shin. Both knees contracted and legs wasted. Ulcers of right arm and hand. Extensive scars on left forearm; large periosteal node on fifth metacarpal. Some wasting of both arms.

This case is known to have had yaws when two years old. Cases of tertiary with history of previous yaws may be multiplied indefinitely.

Among 17 children under the age of 15 years suffering from tertiary syphilis, yaws was admitted in the case of 12. Of the others only two had marks of inherited taint. I regret that I cannot quote a greater number, but one has been so in the habit of expecting the history of syphilis, that enquiry was not often made on this point, and I can only find a reply to this question in the notes of seventeen children.

NON-VENEREAL SYPHILIS

How often do we hear it inferred that a condition cannot be syphilitic because the patient is young and there is no evidence of inheritance. And how often is there a reluctance to attribute syphilis to an adult, from a feeling that this is an unjustifiable slander of his character. Yet thoughtful writers have warned us against this attitude.

> "The physician must forget the local primary lesion, and "put aside the idea that the diagnosis of syphilis carries with "it any stigma of impurity." (Fagge, by Pye Smith.)

I cannot state the truth about the West Indian negro better than by quoting my own words from a report already referred to.

"This applies with far greater cogency in the West Indies, where "the presence of a primary lesion on the genitals of an unmarried "peasant girl implies no immorality. Among the people there is, in "a true sense, no sexual immorality. Coitus among children before "puberty is as common as kissing among European children. The "normal sexual life of the adult is one of transient concubinage, "which does not restrict casual intercourse any more than does the "occasional marriage. The mother of a large family of unknown or "various paternity finds it very tiresome in the daughter of sixteen "years to add another infant to her burden. But that is all. She "accepts the grandchild with simple faith, and tells you that God "sent it."

Four years ago I was asked by the police to see a case suspected to be suffering from smallpox (or Trinidad varioloid varicella). I found the patient, a young woman of decent coloured class, to be a case I had already been treating for syphilis within the last few months.

But the interesting feature was that two little girls, aged about seven and eight years respectively, in the house had now an eruption of papular vesicular syphilides. Not long ago I attended the daughter, aged nine years, of a coloured gentleman. She had been ailing and getting thin for some months, had had frequent and sometimes severe rheumatoid pains, and a persistent rash of papules. She had been under treatment by another physician without improvement. I offered no diagnosis, but prescribed a hundred powders of grey powder and saccharated carbonate of iron. The rash rapidly cleared up, and the general health became robust many weeks before I permitted the discontinuance of the powders. These were instances of non-venereal infection among clean, well-cared for children.

About the same time that the first case above was seen, I was in consultation on a black peasant girl of sixteen years, suffering from a profuse pustular rash, which was thought likely to give rise to a smallpox scare. My colleagues both diagnosed yaws. I said syphilis, and from my point of view we agreed. But one of the others, a man with thirty years' experience of the West Indies, objected—"how could she get syphilis." I have been treating that girl off and on to

the present time for profuse eruptions, iritis, numerous suppurating glands in the neck, bone aches, periostitis. In fact, she has had nearly every early syphilitic symptom possible except nerve syphilis. But she has never had either a venereal chancre or a frambesial eruption.

It is most important that the profession, especially in the tropics, should realise the importance and frequency of extra-genital syphilis. We all know that surgeons and nurses get finger syphilis occasionally. In the island of St. Kitts, with at no time more than eight medical men in practice, and usually only six, there were in twenty-seven years five instances of finger syphilis in surgeons. These are well-authenticated cases.

When we consider how readily syphilis is inoculated, and how the habits of the poor in the tropics afford opportunities for inoculation, we shall not wonder that extra-genital primary occurs. In the West Indies the labourers, especially children, go barefoot, and are, therefore, frequently receiving bruises and scratches on the feet and legs.

The negroes never realise the fact of infection or contagion. This forces itself on our attention by the trouble they give when quarantine measures are to be carried out. They are too indolent or careless to make any effort to avoid or prevent infection, even when they acknowledge that a disease is catching. A child with yaws huddles together with the other children just as naturally as the adults will sleep or cohabit with lepers. In Tortola, while I was there, the only female who could be called a professional prostitute was an ulcerated leper. The syphilitic, the yawsy, the leprous and the healthy mix in free and intimate contact without a thought. Leprosy, like elephantiasis, may be the subject of reproach, but these diseases are not a cause for avoidance of physical contact. Yaws is hardly even a disgrace, except in the eyes of schoolmasters and other coloured folk of a better class than the labourers.

Under such circumstances, it is hardly wonderful that yaws can be extremely prevalent in a native community; and it would be miraculous if non-venereal infection did not occur where syphilis is almost universal.

I think observers in other longitudes, who see the people, not in hospitals, but in their huts, will find my description of West Indian

peasant life not unfamiliar, and if a look-out is kept, I am sure others will verify the existence of non-venereal syphilis for themselves. In places where yaws, under its various names, is prevalent, it will be found that frambesial eruptions and non-venereal syphilis of ordinary type are running together. In the drier and less-wooded localities the latter will probably be found the more common form.

It must be remembered that in inherited syphilis secondaries do not occur after the second year, and are usually over in the first year of life. Tertiaries are the only lesions by which inherited taint can be manifested in children of, say, three years and upwards. Jonathan Hutchinson lays down :—

" If secondary symptoms of the kind described are to occur at all, " they will show themselves in infancy, and in the vast majority of " cases within the first three months of life. This is a very important " fact. If a syphilitic infant survive the first outbreak, in the course " of from six months to a year the symptoms common to this stage " (the rash, snuffles, mucous patches, &c.) will wholly disappear, and " there will follow a period of some years during which no active " symptoms will occur."[1]

When, therefore, undoubted secondary eruptions and swollen glands are seen in children over the age of two years, they are almost certainly due to acquired disease, and this certainly rapidly increases with the age. Such children should be examined for the primary, which in the bare-footed is usually on the foot, most commonly on the outer malleolus. The indurated pigmented scar may be seen some time after the sore has healed, but these sores are often of long duration, and may be still unhealed months after they were infected with syphilis. They are often already indolent ulcers before the infection. I have diagnosed such primaries before the general symptoms were manifest, and the later eruptions have justified my diagnosis. More usually, however, one passes the sore by until the patient is complaining of rheumatoid pains or fever, for sore feet in children are so common that we cannot treat them all as potentially syphilitic.

For several years I have been impressed with the frequency of these extra-genital chancres in children, followed by secondaries of

1. Syphilis, Ed. 1901, p. 75.

the ordinary type, and some of my colleagues have adopted or confirmed my opinion. Such cases are seen more commonly in towns where yaws is never prevalent. I have seen a few such develop frambesial eruptions after having been diagnosed and treated as syphilis; the frambesiae appearing along with the other syphilides.

Nearly everyone familiar with yaws recognises what the natives all know, that there is a primary in yaws. No doubt this is often a fungating ulcer rather like a frambesia, but it may be identical with the foot chancre of ordinary type. Chancres on the penis not infrequently fungate in the same way, as Hutchinson points out. The foot chancre, out of which yaws also sometimes originates, is, in my experience, a small sore about 5 to 10 mm. wide, round, with indurated base and periphery and raised edges. It may be on one of the toes, the inner malleolus, the tubercle of the fifth metatarsal or back of the foot, but the commonest site is the outer malleolus.

In the case of yaws, the natives in some instances do not recognise the primary, which may be inconspicuous, as is often the case in syphilis; and then a large group of frambesiae, especially a horse-shoe group, may be called the "mother-yaw." On the other hand, a primary may really become the "master-yaw" or largest frambesia, by fungating and having a satellite eruption of papillomata around it.

It is perhaps unnecessary to insist that there is aways a primary in yaws. Numa Rat has made this quite clear, and the universal opinion of the inhabitants of yaws districts themselves leaves no doubt on the subject. Notwithstanding, it is urged (e.g. Manson, Tropical Diseases, p. 530) as an argument against the syphilis theory that the primary is "wanting in yaws."

The identity of non-venereal syphilis of the easily-recognised type, such as I first referred to, and the whole group of symptoms called "yaws," is so obvious that the peasants themselves bear witness to it. They include under this name other skin eruptions, besides the frambesiae, which they have observed to be associated with the latter. These have popular names—tubbo, corn-yaws, crab-yaws, pian-gratelle and dartres. The peasants speak familiarly of "yaws-pains." They point to where a patient did "ketch a yaws," who has never developed frambesiae, but had rheumatoid pains and vesicular syphilides, and is afterwards seen with tertiary.

It is well to consider some of these other yaws eruptions, which are so well recognised by the natives and described by writers.

Tubbo is a papilloma, pathologically the same as the frambesia, though unlike it in appearance, which breaks through the thickened skin of the sole, and is modified by the pressure of the hard epidermis, in the same way as the small papules of early psoriasis are modified when they appear under the skin of the palm.

Corn yaws in English-speaking colonies covers all discrete papular, vesicular, and even pustulo-vesicular eruptions.

Crab yaws is simply palmar and plantar psoriasis. It is sometimes severe, and becomes a moist dermatitis. More usually it is seen some time after the yaws, and is identical with the tertiary plantar psoriasis. It is chronic, and very resistant to treatment.

Pian gratelle, as described by Numa Rat, Alford Nicholls and others, is obviously the small papula of syphilis-lichen. It appears at an early stage in a profuse crop usually, and, as in European syphilis, it may reappear as a late secondary, and its presence indicates a severe infection. This tendency to recurrence and this significance of the papular eruption is insisted on by writers on both syphilis and yaws (cf. Jamieson, Diseases of the Skin, p. 544, and Alford Nicholls, Report, p. 285).

Dartre (or as Nicholls calls it, the macula or squama) may be perhaps in some instances the macular rash, which is the earliest skin symptom of syphilis. The colour of it cannot be seen on a black skin, but when severe and on unwashed skin it may possibly result in an exfoliation of the corneous layer of the epidermis; but as described it is the syphilitic scaly psoriasis. This is the earliest eruption in yaws, and is very common. It is frequently coincident with the papular eruption—lichen—when that is present, and sometimes lasts to the stage of the frambesial eruption. Its appearance is absolutely characteristic of scaly psoriasis.

All these obviously syphilitic conditions are recognised by the natives to be essentially parts of yaws as a disease. A patient is said to have yaws on the strength of any of these, even by medical men, without the frambesial eruption.

Alford Nicholls and Numa Rat, who have made long and careful observations of yaws in its earlier as well as later post-frambesial stages, describe these conditions as belonging to yaws. Dartre and

pian gratelle especially are noted by them as being characteristic of yaws. In my own experience, psoriasis and lichen are just as frequent in occurrence in cases which develop frambesiae as in those which do not. The rheumatoid pains are almost always a feature of yaws, and these, as well as the less frequent palmar and plantar psoriasis and vesicular syphilides, are identical in both diseases.

In short, the *vox populi* has declared the truth, which the medical profession has not yet generally accepted, that yaws is non-venereal (or sometimes venereal) syphilis of the tropics. At any rate, the people name syphilitic manifestations and associate them with the frambesial eruption under the one name yaws.

A few recent writers on yaws, under its various names, have seen and recorded these facts; proving that yaws is more than the eruption of frambesiae, as described in text-books (e.g. Scheube). Even those writers who have not included these symptoms as part of the same disease have noted the frequency of the occurrence of syphilitic manifestations together with frambesiae. They regard the smaller syphilides as evidence of syphilis and reserve the frambesial eruption to be called yaws, and then speak of the concurrence of the two diseases. Daniels and Wallbridge note that "a fair number of such cases are "recorded, and in all the syphilis preceded the yaws."[1] It is doubtful whether by "syphilis" they refer to primary syphilis, i.e. the genital chancre from their point of view, or to small syphilides. Blanc, of Tobago, observes:—"I have not seen any yaws patients with syphilis "in the primary or early secondary stages, but have very commonly "seen them in the early stages of yaws with well-marked papular "eruptions, which, however, might have been either syphilitic or the "precursors of yaws tubercules. These symptoms commonly "disappear under treatment, but sometimes give rise to a scaly "eruption very like psoriasis. . . . Syphilis, as evinced by severe "cachexia, or indurated and enlarged glands, enlargements of bones, "nodes, &c., has been a very frequent complication in the cases of "yaws."[2]

There can be no doubt that the other secondary manifestations of yaws are identical with symptoms elsewhere ascribed to syphilis. Some writers attribute them to yaws, others to a concurrent syphilis.

1. J. Hutchinson, Fasciculus, p. 19.
2. Nicholls' Report, p. 165.

But anyone who cares to look for them will not have far to seek in almost any case of yaws.

Consistently with the description of the secondaries of yaws, Rat goes further and describes the tertiaries. In this he is in accord with Daniels and Corney in Fiji. These symptoms need no description—periostitic nodes, multiple dactylitis, destructive rhinopharyngitis, lupoid ulcerations, &c. It is only to be remarked that one does not realise to what extent tertiary syphilis can go until he has seen the cases of untreated syphilis in a yaws district.

It is evident that, in the experience of the writers just mentioned, there is not enough venereal syphilis to account for the extraordinary prevalence of tertiary, and so they were driven to ascribing the tertiary, quite correctly, to yaws. They have failed, no doubt, to see the non-venereal syphilis without frambesiae, which would have afforded an escape from the difficulty.

Tertiaries identical with those of syphilis are recorded as being exceedingly prevalent, and are attributed to yaws by Daniels and others in Fiji, Kynsey in Ceylon, Rat in Dominica. Among ninety cases of tertiary syphilis of whom the history was ascertained in St. Vincent, sixty-nine were said to have had yaws and only five admitted genital syphilis. Some of the remainder had suffered, no doubt, from extra-genital chancre without frambesiae, and therefore did not recognise that they had yaws. On the other hand, not all that claim to have had yaws have had a frambesial eruption, for, as I pointed out before, the natives, and apparently also some medical men, call cases yaws that have had no frambesiae.

From the facts of the polymorphism of the eruptions and the occurrence of tertiaries, it is evident that the only difficulty in the way of the general abandonment of the yaws heresy is the frambesial eruption itself. As Jonathan Hutchinson has pointed out, the profession did not originally hold yaws to be an independent disease, so that it is quite fair to speak of this belief that yaws is not syphilis as a heresy. The frambesial eruption is of such importance and, to my mind, such a formidable difficulty that I have reserved its consideration for another section.

My contention is that syphilis is very common, in fact, almost universal, among the natives in the tropics; that in certain districts it usually presents an eruption of papillomata, which has given rise to

the idea of yaws as a distinct disease; but that the cases with papillomata present otherwise all the features of syphilis, just as do the non-venereal cases. To appreciate the relation of the various forms of eruptions to each other and to recognise their nature, as well as to understand many other disease problems, it is necessary to remember that syphilis is in the tropics not usually a venereal disease. Notwithstanding the vitality of long-rooted error, I feel sure that the profession would have more readily accepted the dictum of Jonathan Hutchinson had we realised the frequency of extra-genital chancre. It is the teaching of the schools and text-books that syphilis is a venereal disease, which has blinded us to the fact that in the tropics it has little to do with sexual intercourse; that there is, in fact, far more non-venereal syphilis in the world than syphilis acquired on the genitals.

CASE 14.—D. K.; 6 years; Coloured.

March 27th, 1906. Has had a sore of the toe and paronychia for several weeks. About two weeks ago began to get "spots" on the skin. Has now a few spots of psoriasis over the body and palms of the hands. Also on the knees and elbows. Is feverish at nights, has small hard glands in both groins and on elbows.

Here is a very early case in which, I think, the diagnosis of syphilis is justified.

CASE 15.—J. W.; 13 years; Black.

August 1st, 1903. Has a small indolent dirty sore on the left outer malleolus, with raised edges and indurated base and periphery. This has been present for several weeks. There are tender glands in both groins. That on right larger and more tender. Small hard glands left side of neck, on both elbows and in both axillæ. Two weeks ago the first crop of eruptions appeared and they have continued to come out since. Has now a general papular eruption (lichen) tending to vesicular heads. This is very closely set over the whole trunk, extending to the neck, upper arms, and thighs, where it thins away. A few papules on the lower part of the cheeks, none on the limbs.

Also an early case, in which the diagnosis seems very evident.

CASE 16.— R. B.; 10 years; Black.

April 6th, 1903. Three months ago had a scratch on right outer malleolus which became a sore. Her mother shows this as what she considered a mother yaw. It is now a pigmented and indurated scar. There are many spots of scaly psoriasis over the body, face and limbs—rheumatoid pains in the right elbow, both knees and ankles. The mother complains that the child is getting lazy and hard to rouse in the morning. Later she developed evening fever.

September 1st, 1903. Noted that she had attended irregularly. Eruptions are worse. Has never had frambesiae.

This illustrates the popular identification of syphilis with yaws.

CASE 17.—M. N.; 14 years; Coloured.

April 22nd, 1903. Has a small excavated ulcer about 6 mm. wide on a raised indurated base, on the outer side of the right foot. This has been present for several months. Hard enlarged glands in the right groin. Swollen tender glands in the right side of the neck. A chain of small tender glands in left side of neck. Fever, malaise.

A patch of superficial flat large papules with crusted scabs, each 10 mm. in diameter, on the front of the right thigh. These are like mild rupia or ecthyma.

CASE 18.—L. P.; 21 years; Black.

April 2nd, 1903. Has a deep excavated ulcer on the left outer malleolus. Does not admit venereal primary. Has no leucorrhoea to indicate cervical chancre. A coarse papular desquamating eruption of lichen on both hands and feet. Few papules on the face. Very profuse vesicular eruption on both legs and thighs. Glands enlarged in both groins and on both elbows.

In spite of the age of the patient, I had no hesitation in attributing her syphilis to infection on the ankle.

CASE 19.—S. N.; 30 years; Black.

March 15th, 1903. Has four small ulcers with indurated bases and raised, thickened, everted and irregular edges, round the right nipple. These appeared first three months ago while she was suckling an infant with yaws. They have all the appearance of primary chancres.

On 6th May, 1903. Complains of pain all over the body, malaise, and constipation. Looks and says she is very ill. Has nervous sensations and sleeplessness.

She never developed any eruption. Continued ailing and sickly for at least two years under treatment for which she attended irregularly, as she felt better or worse. Treated with small doses of mercury from the first. Never salivated. At length she complained mostly of gastric troubles. These were not due to mercury, for she only took doses of 10 minims of liq. hydr. perch. three times a day. After two years recovered health.

This was a case of visceral syphilis contracted from yaws. I have now under treatment a very severe case of gastric syphilis, in which the gastritis followed a year after a genital chancre. In this, also, there was no eruption, but adenitis and iritis.

CASE 20.—I. D.; 9 years; Black.

February 25th, 1904. A wide ulcer on the left outer malleolus, with a dirty yellow crust. Profuse patches of psoriasis and areas of lichen over legs, trunk and arms.

This would be called an early case of yaws, with the maman-pian, dartre and pian gratelle. By changing these names it becomes syphilis.

CASE 21.—G. P.; 9 years; Black.

1906. Small punched ulcer about 5 cm. wide on the back of the foot near the toes, with raised thickened edges. This has been present for several months.

There are hard glands in both groins, large soft glands in the neck. History of "glass-pox" in successive crops for several weeks, some months ago. Now comes complaining of fever, malaise, and anaemia.

In this case the knowledge of the occurrence of non-venereal syphilis led one to inquire about the eruption. This, together with the glands, the primary and the cachexia, leave no doubt as to the diagnosis. Chicken-pox, of course, does not continue in crops for several weeks.

CASE 22.—V. T.; 4 years; Black.

Mother was treated for a copious syphilitic rash eighteen months before. The patient was treated for an eruption some months ago. Now has a papular rash (lichen) over neck and shoulders. There are patches of weeping and exfoliating dermatitis over each side of forehead, on both scapulae, and on back of shoulders.

This is too old for hereditary syphilis. It is not unreasonable to attribute the rashes to syphilis acquired from the mother.

CASE 23.—R. S.; 5 years; Coloured.

1905. Multiple ecthymatous sores encrusted with yellow scabs on the legs. Large patch of the same on the left thigh, and a flat condylomatous spot with dry yellow crust among the ecthymata. Pigmented marks of large pustules about the body generally. Keratitis of one eye. Treated with mercury.

December 20th, 1905. Eruptions have disappeared, leaving no scars, but pigmented stains which subsequently faded. Admitted to Hospital for corneal ulcer and treated till well with mercury internally.

Here, as in the last case, there was no primary to be found, but the condition was evidently syphilitic, the age too late for secondaries of inherited taint. The eruption was one like frambesia, and even presented one spot of condyloma. The crusts, however, did not cover hypertrophied papillae, but eroded spots.

CASE 24.—M. D.; 3 years; Black. Mother seen on 28th October, 1902, with profuse eruptions of circular patches of scaly psoriasis over whole body, limbs and face.

January 5th, 1904. The child has a multiple eruption over both arms and legs, and back, of scaly psoriasis; patches of lichen in spots; and circinate and exfoliating dermatitis.

The age is too late for the secondaries of inherited syphilis; besides the mother has acquired syphilis only about fourteen months before. If the child had inherited syphilis from the father the mother would have been immune, by Colles' law. It is clearly an instance of acquisition from the mother.

CASE 25.—N. P.; 8 years; Black.

March 29th, 1903. Ulcer, 50 mm. wide on the dorsum of the left foot, which originated from a scratch three months ago. This has a raised fungating surface, and thickened raised everted edges. A very large gland in the left groin, large gland in right groin, gland in left elbow, and chains of shotty glands in the neck. Has intermittent fever, and constipation with dry hard stools. There is a desquamating condition of the skin of the extensor surfaces of forearms and legs, and a patch of coarse papules like common psoriasis on each elbow.

July 28th, 1903. Foot has healed. A flat yellow crusted eruption is on both knees and both elbows. Dry crusted warts on both hands.

This may almost be given as a case of yaws, except that the eruption was not perfectly frambesial. The patient lived in town; had he lived in a country village he would, no doubt, have had yaws.

These few cases, I think, illustrate the various aspects in which non-venereal syphilis may be presented to the physician: primary syphilis not followed by frambesia contracted from yaws; secondaries in children with and without an evident primary; secondary in an adult due to extra-genital infection. Two show the difficulty there may be in differentiating yaws. I selected five which were seen within five weeks at a time when my notes were being regularly kept. This number indicates how common such cases really are. (Cases 16, 17, 18, 19 and 25.)

ATTITUDE OF THE MEDICAL PROFESSION TOWARDS YAWS

In consideration of the symptoms of yaws, as referred to above, there would certainly never have been a doubt as to its identity with syphilis had it not been for the frambesial eruption. This is so unusual in European syphilis, and such a distinct and characteristic manifestation in yaws, that it was natural it should overshadow the other symptoms. Most physicians, who have not looked carefully into the matter, have a picture of yaws as consisting only of the frambesial eruption; they think this is a harmless, though unpleasant, disease, which usually runs its course and disappears spontaneously, without permanent injury to the health. But at first yaws was included with syphilis, and even since its separation as a disease *sui generis*, the doubt that it was correctly separated has existed.

In view of the enormous prevalence of yaws in the tropical world, this doubt gives an importance to the disease which, I think, has not been appreciated; for it is a most serious matter if syphilis, the most destructive and far-reaching in its effects of all diseases, is being allowed to ravage whole communities untreated and almost unnoticed.

When in recent years the syphilist, who for forty years has been the authority to the English-speaking profession, pronounces his unqualified opinion that syphilis is yaws, it is time that the world took up the matter seriously. In his preface to Numa Rat's work on yaws, Jonathan Hutchinson expressed this view, based, it appears, largely

on the evidence offered by Rat himself. Since then, after further investigation, he has delivered, so to speak, an *ex cathedrâ* opinion to the same effect in Fasciculus XIV of the new Sydenham Society's Atlas, and presented the evidence for his opinion. Notwithstanding, the medical profession, with few exceptions, are in the same attitude of indifference. The new student of tropical medicine will hardly get from his text-books any indication that there is any serious or reasonable doubt about the independence of yaws. The 1903 edition of Manson makes a misleading reference to Hutchinson as holding "that yaws is possibly syphilis modified." The English edition of Scheube, 1903, only refers to Hutchinson to misunderstand him in a similar way.

Somewhat in consequence of this, those who believe in the identity of syphilis and yaws are in a small minority. Not many take much account of yaws at all. Of those who believe in the independence of yaws, it is only the few enthusiasts who have studied the matter that think it a serious disease. It is not to be wondered at therefore, that in most places yaws is not treated at all. Trinidad, especially in the island of Tobago, is the only colony in which I have heard of any rational and systematic attempt to deal with the disease, but even there, because they do not realise its gravity and true nature, the measures taken by the medical officers must be incomplete. In some other places the yawsy are in effect regarded as social offenders, and forcibly removed to yaws hospitals for treatment. Such a course can never obtain the sympathy and support of the people whom it is meant to benefit.

In the treatment at all yaws asylums and dispensaries, as far as one can judge from the literature of the subject and the reports, the frambesial eruption is regarded as the disease. When this dries, the patient is said to be "cured"; when another crop appears this is called a relapse. All physicians that have given mercury find that the eruption disappears rapidly as a rule, and that it is apt to recur when the mercury is stopped. Some think that the mercury has a bad effect, and that the yaws should better be allowed to run its course. Certainly the frambesiae disappear under the mercurial treatment, and as certainly they sometimes recur. No one thinks he has "cured" a case of syphilis because a crop of secondaries has faded; nor does he stop mercury on that account; nor is he surprised if more

secondaries appear even while his patient is still taking mercury. When treating a case of syphilis, one is thinking not of the secondaries but of the tertiaries. The second stage will usually run its course and the patient get well in time without mercury. But it is, as a rule, only the mercury given early that saves the patient from a life of tertiaries later on.

If the papillomata are all that there is, then yaws is hardly worth much trouble; but the observations, especially of Numa Rat and Daniels, have established the fact that there is an aftermath of evils. They could not help but attribute to the effects of yaws the vast amount of exaggerated ulceration, periostitis, necrosis, and dactylitis which they saw around them. Numa Rat boldly calls these the tertiaries of yaws, though he still believes yaws to be distinct from syphilis. If the diseases are different, then yaws is the worse of the two, and it is the most terrible scourge from which the human race suffers.

If even yaws is not syphilis, then there is still abundant indication that the treatment is the same, and we should insist on the early and persevering use of mercury; for the time for treatment of tertiary syphilis is in the early secondary stage, and by inference the same must be true of yaws.

If, however, we decided to regard yaws as syphilis, its importance would be at once recognised by every member of the profession; and the people, educated in time to pay proper attention to the disease, as Europeans do to syphilis, would assist in the effort to limit its spread and cut short its dire results.

A systematic and conscientious treatment of the cases, and a persistent education of public opinion, would at length effect what all the yaws asylums, dispensaries and commissions have failed even to begin.

In view of the theory of Hutchinson that yaws is the original disease, we may abandon the name syphilis, which bears but an indifferent repute, and speak of yaws. Recognising its nature, we may teach the prophylaxis of syphilis under its various local names. The writer has already prepared, with this view, a primer of hygiene for West Indian negro children, at the request of the Board of Education of St. Vincent.

I said before, that it is only the frambesial eruption which constitutes the difficulty in accepting yaws as syphilis.

Manson says:—"I may mention the primary sore, the infection "of the foetus, the adenitis, the exanthem, the alopecia, the absence "of itching, the iritis, the affection of the permanent teeth, the bone "and eye affections, the congenital lesions, the polymorphism of the "eruptions, the nerve lesions, and the gummata of syphilis. All these "are wanting in yaws."[1]

To take these in detail:—

(1.) The primary sore.—There is, as we have seen above, no doubt at all about this. Numa Rat made this quite clear, and the experiments of Charlouis are convincing. In short, as Hutchinson says, Alford Nicholls is the only observer who disputes the fact that there is always a primary lesion in yaws. Careful examination and enquiry will elicit this quite as often as in cases of syphilis.

(2.) The infection of the foetus.—It would be quite as logical to deny the inheritance of syphilis. It is so rare for a child to be born with syphilitic eruptions that few of us have seen such a case. Many infants are seen with yaws at the age that they might have the secondaries of inherited disease. But when such cases occur, we are in the habit of inferring that the yaws is acquired. Inheritance has been demonstrated in at least two cases mentioned by Hutchinson. I have never myself seen an infant known to have hereditary syphilis develop a frambesial eruption; but all the infants I have seen thus have been at once put on mercury, and kept at it as long as I continued to see them. They get better, or, more usually, I hear nothing of them for a long time until the parent applies for a death certificate. It cannot certainly be said that inherited syphilis is not sometimes frambesial, and it is known that a child may inherit the yawsy taint and develop the frambesia in due time.[2]

(3.) The adenitis.—This is an almost invariable accompaniment of the yaws infection. The experiments of Charlouis are conclusive, besides the experience of actual examination of yaws patients.

(4.) The alopecia.—In all my experience of many hundreds of cases of negro syphilis (non-frambesial) I have only once seen alopecia, and in that case the diagnosis was at first obscure, but

1. Tropical Diseases, page 530.
2. Hutchinson Fasc., page 18.

afterwards cleared up. A physician of far longer experience than mine of the West Indies tells me he has never seen syphilitic alopecia in a black patient.

(5.) The absence of itching.—This is admitted to be by no means an absolute rule in syphilis. Nor is itching at all the rule in yaws. When we consider the irritation of the small flies around a yawsy patient, the admission of itching in some cases is not remarkable.

(6.) The exanthem.—It is difficult to understand which particular eruption is referred to. All or most of the syphilides have been seen in yaws.

(7.) The iritis.—This is distinctly uncommon in negro syphilis, and is a comparatively rare symptom anywhere. It is quite feasible to imagine that, where syphilis is profusely manifested on the skin, the deeper structures, particularly the nerve system and its appendages, may escape.

(8.) The affection of the permanent teeth.—This must be extremely rare, at any rate, among West Indian negroes. I cannot remember ever having seen notched incisors in a black person. Finucane and Corney, as mentioned by Hutchinson, report having seen such occasionally in Fiji, where syphilis is not admitted to be present. One has the impression that either the negro race is exempt from this affection, or that it is caused by mercury given in infancy. I very frequently look at patients' teeth to note the tolerance to mercury, and also to note the irregularities which constitute a stigma of degeneration.

(9.) The bone affections.—These have been described frequently as tertiaries of yaws. But the author has already denied them to us by saying that they are due to an independent intercurrent syphilis.

(10.) The congenital lesion.—There are none in syphilis, except the rare pemphigus neonatorum, and the syphilitic causes of still-birth. No one has attempted to prove that miscarriage is any less frequent among yawsy mothers than among the syphilitic.

(11.) The polymorphism of the eruptions.—This is abundantly evident in yaws, and is referred to by Manson himself.

(12.) The nerve lesions and the gummata of syphilis.—The negro race is not prone to suffer from nerve syphilis, but all tertiaries and parasyphilis are taken from us by the simple assumption of no intercurrent syphilis, of which there may be no other evidence.

As far, then, as all these differentia are concerned, it is not possible to separate the diseases. It is impossible to overlook the tertiary stage of yaws. The indications for treatment are the same as for syphilis. Even if the "syphilis theory" is not true, it is *as if true*, as Paul Carus would say; and, therefore, it is good to teach and useful to believe. Any abstruse difference based upon the frambesial eruption can only have an academic interest.

THE FRAMBESIA OR PAPILLOMATOUS SYPHILIDE

The frambesia has been variously referred to by writers as a tubercle, a sore, granuloma, or a papule. No doubt, it is not of an invariable form, but none of these terms are correct. In every case the frambesia is raised, and consists of a closely-set group of hypertrophied papillae. The dermis is not ulcerated, for properly the eruption leaves no scar. The frambesia is sometimes naked and moist, but more usually it is covered by a yellow crust. In some instances, and especially when the eruption is old, the crust is dark or dirty brown, but typically it should be yellow. There may be a yellow secretion under the crust, and in the early and active stages this is the rule. When old and undergoing resolution the frambesia is dry, and the crust, if present, is more adherent. If the yaw has been naked, it becomes in the dry stage covered with a film.

Usually the eruption disappears by slow absorption, each yaw becoming imperceptibly smaller day by day. No doubt, yaws occasionally ulcerates by accident or by bad treatment, and leaves a true scar, but this is by no means its natural tendency.

As a rule, the frambesiae are scattered more or less profusely over the skin. The corners of the mouth, around the anus and on the genitals are the favourite sites. Typically each yaw is round or oval, and measures 8 to 15 mm. across. But they vary much in size, and may be much larger. They are sometimes arranged in a circinate manner, and by coalescence of contiguous frambesiae a large, irregular or horse-shoe form may be produced. It is, certainly, a very striking thing to witness the uniformity of the appearance of a number of cases. One can go to a country village in a yaws district, as in most parts of St. Vincent, and collect twenty children in five

minutes all showing the eruption in almost identical form. There is small wonder that the profession, having regard to this eruption alone, has almost generally fallen into the way of making it constitute a distinct disease.

Alford Nicholls, who has made a thorough anatomical examination by sections of the frambesial as well as of the papular and "macular" eruptions of yaws (fig. XIX, in his report), shows that the papule of pian gratelle consists of a few hypertrophied papillae infiltrated with small round cells, which are also accumulated in the underlying dermis; the corneous layer of the epidermis is undergoing overgrowth and exfoliation. This is identical with the structure of a papule of syphilitic lichen. (The frambesia is beautifully shown in section in fig. XXII.) It consists entirely of very much elongated and hypertrophied papillae, with a great infiltration of round cells. The epidermis is desquamated so that the ends of the papillae are covered only by the secretion and the crust, which is formed of layers of dry secretion. The papillae are closely crowded together. This structure, without the crust, is identical with that of the syphilitic condyloma. The frambesia is, in fact, one of the large papules of syphilis. It is not, therefore, a granuloma in the sense that a gumma is. The small round cells are certainly the elements of granulation tissue, but the whole mass retains its physiological character as a group of natural papillae, which do not disappear so as to leave a scar. They have undergone an evolution and hypertrophy, and will return to their normal size by involution and absorption of the added elements. The term granuloma is a misleading one. The red granular surface of the frambesia, seen after the removal of the crust, to which it owes its name, from the resemblance to a raspberry, is not due to granulations such as are on an ulcer.

The proper pathological description of the frambesia is in the term papilloma; and it is structurally identical with the papillomata of syphilis, condylomata and rhagades. In a drying papilloma the crust fits into the interstices between the papillae, so as to be difficult to remove. But when it is removed, the individuality and tough consistence of the papillae can be easily demonstrated.

On moist surfaces, as on the vulva and around the arms, and sometimes at the webs of the fingers and toes, the papilloma is naked. The ends of the papillae are swollen, and squeezed together so as to

give a flat surface. Such frambesiae are indistinguishable from ordinary condylomata.

We have seen that it is only in the frambesial eruption that yaws differs from syphilis, but even the frambesia is identical with the syphilitic papilloma on a moist surface.

There is left, therefore, only the occurrence of condylomata all over the skin which can be held to be distinctive of yaws. Jonathan Hutchinson points out that a general papillomatous (frambesiform) eruption does occasionally occur in Europe. It has even been suggested that "Sibbens" was a syphilide of this form.

It is quite feasible to argue that on the thick, highly-developed and active skin of the dark races this exuberant form of eruption takes the place of the milder papules known in Europe. The higher infectiveness and greater prevalence of the frambesial syphilis is readily explained. Syphilis ordinarily (that is, as known among whites) is contracted from the primary and from moist papillomata-mucous plaques and condylomata. Yaws, however, has so many of the moist infective lesions on the surface of the body that contagion is far more easily effected under any conditions of life; and in the circumstances, and by reason of the habits of the natives, it must be almost impossible for each patient not to infect someone else. Infection can, of course, be effected venereally. Dr. Hatton, of Grenada, says, "I have known cases occur from sexual connection."[1] Dr. Boyd, of Grenada, reports a case, presumably of venereal infection of yaws:—"J. W.; a Creole. At 16 years, was perfectly healthy and "unexposed to any source of contagion, until deflowered by a man in "March, 1891. Fourteen days after this she found some tubercles on "the vulva and inside of the thigh, for which I admitted her to the "yaws hospital."[2] There is no suggestion that the man had yaws, and the appearance of a secondary on the inside of the thigh so soon after infection renders the case unreliable. Neither secondary syphilis nor yaws "tubercles" appear until some while after the primary. It is possible that the so-called tubercles were all small primary sores, those on the thighs being inoculated in finger-nail scratches received during the struggle.

1. Nicholls' Report, p. 178.
2. Nicholls' Report, p. 198.

It is a fact that yaws is far more common than syphilis in the villages, and in towns *vice versâ*. When a medical man sees yaws in an adult, he never thinks of enquiring as to the possibility of its having been acquired venereally, and when he sees a case of ordinary syphilis he assumes the venereal infection, in spite, perhaps, of the asseverations and protests of the patient. In the country districts of yaws countries the syphilis, however acquired, probably for the most part shows frambesiae; while in the towns the non-venereal infection gives rise to the better-recognised forms of syphilis associated in our minds with venereal infection. Dr. Tulloch, of Tobago, remarks that the "primary sore is especially rare in the outlying districts where yaws is most common."

More observations are wanted on two points:—

(1.) Syphilis (without frambesiae) contracted from yaws.

(2.) Yaws (with frambesiae) contracted from ordinary syphilis.

As the date of infection will be three weeks or more, in both cases, before the first manifestation of the primary, and usually two or three months before the appearance of the secondaries, it will be extremely difficult to get histories, especially from ignorant peasants, to illustrate these two points. I have already described one instance of syphilis contracted from yaws (Case 19). Some cases are available of yaws supervening on venereal chancre, but the history of these does not include any proof of the nature of the disease from which the women suffered before they transmitted it as a venereal chancre.

I shall describe one such case. Similar cases, though not interpreted as I do, are reported by Numa Rat, W. Boyd and H. Bennett in the West Indies, and by Wallbridge in Fiji.

Rat's case is as follows:—"A man consulted the doctor about an "indurated sore of the prepuce, and had at the same time a small "ulcer on the ankle. During two months he was under mercurial "treatment, and had in this time sore throat, roseola, psoriasis and "enlarged glands. He then noticed a change in the ulcer; the "graulations became pale and large. Three weeks after this the "ulcer healed. A week, again, after this came a wide-spread crop of "papules. A few of these developed into frambesiae. The "'syphilitic roseola co-existed with the yaws papules, and the

"'syphilitic psoriasis with the yaws tubercles.'[1] A clearer case of "frambesial eruption supervening on venereal infection of syphilis "cannot be found."

W. Boyd's case is given as follows:—"A man had a well-marked "and indolent Hunterian chancre, and bubo, when he came under my "notice, and at the same time a mild eruption of frambesial tubercles, "which he stated made their appearance two weeks after the "syphilitic sore."[2]

H. Bennett's case:—"The patient was a young woman, who "contracted the syphilis first, and then got either a primary or a "secondary ulcer inoculated in an unknown manner with the yaws."[2] From the confidence with which the syphilis is spoken of, one presumes that a venereal chancre was known. I have, therefore, included the case here.

Dr. Wallbridge's case is quoted by J. Hutchinson[3]:—"The "patient, a white man, had syphilis, and developed frambesiae. The "latter disappeared on his return to England, and later, after he "again went to Fiji, he suffered with tertiaries."

In all these cases it is, of course, impossible to prove that the persons from whom the disease was contracted had syphilis of ordinary type.

There still remains one fact which forms the strongest objection to the identification of yaws with syphilis. This, so far as I know, has not been referred to by previous writers, and it remains for one, who for the last twelve years has been holding and teaching the syphilis theory, to raise what seems to be the strongest argument against that theory. Although it may be true that the nature of the dark skin predisposes to the frambesial eruption, it is indisputable that yaws is seen breeding true on white persons. In a yawsy village the poor whites get the disease and have the eruption just as certainly as do the blacks. On a white skin the frambesiae are less developed. The papilloma is not so prominent, and the crust less firm. That is, the yaws is flatter, moister, and usually smaller than on a black patient, and it is associated with more of other syphilides, psoriasis, large moist papules, ecthyma, &c. It is a condyloma

1. Nicholls' Report, p. 168.
2. Nicholls' Report, p. 166.
3. Fasciculus, p. 19.

of skin, nevertheless, and is of frequent occurrence; while in Europe such a lesion is so rare as to be a curiosity.

In considering a probable explanation of this phenomenon, we must note that even among negroes the frambesial eruption is rarely seen in towns, and while it is common in some West Indian islands, it is rare or absent from others.

I have already suggested that damp, hot places in the tropics, with dense vegetation, show a prevalence of yaws. This, I think, will be found a rule. Numa Rat states that yaws is absent from Anguilla,[1] which is flat, dry and barren. The typical yaws district is a deep valley or ravine in a tropical volcanic island, where vegetation is rank and the rainfall high.

Yaws is, therefore, very prevalent in Dominica, St. Vincent, St. Lucia and Grenada. It occurs to a less extent in Antigua, St. Kitts and Nevis. Antigua is flat and not wooded, except at one part. St. Kitts and Nevis, though lofty and volcanic, have long slopes sweeping away to the sea. The land here is cleared and cultivated. There are a few bushy ravines with villages, and in these yaws may be found.

There is, then, another factor besides the character of the skin which determines the appearance of frambesiae. We cannot yet predicate this factor, any more than we can explain many other examples of the variability of syphilis. Why, for instance, does lupoid ulceration of the face happen so frequently on the Windward side of St. Vincent, while destructive rhinopharyngitis is the rule on the Leeward side? Why do some patients get a formative periostitis, and others necrosis of bone? I may be allowed to copy from a recent letter of mine[2] :—"To understand this protean disease, "we must realise that various factors intervene to determine its "manifestations. We can appreciate some of these. The races and "individuals who tax their nerve system suffer from nerve syphilis, "which is comparatively rare among negroes. The negro, with his "highly-specialised and active skin, displays an exuberance of "eruptions rarely seen in Europe. The labourer, exposed to injuries, "develops grave bone lesions. The women in St. Kitts, who

1. Report on Anguilla. Colonial Reprints, No. 20.
2. Journal of Tropical Medicine, May 15th, 1906.

" habitually gratify the desire of sailors for paedicatio mulierum, get " stricture of the rectum. Some of the determining factors we have " no knowledge of, others we can now guess at."

The frambesial eruption, therefore, though a striking and characteristic feature of yaws, we have seen is not unknown in syphilis. The difference between yaws and syphilis is only in the frequency of occurrence of this eruption. If all that is called so is yaws, it must be admitted that the frambesia is not even an essential of yaws. In face of all the other points of identification, it is impossible to give this one feature such pre-eminence as to exclude the others. We must accept the papillomatous eruption only as a phenomenon of syphilis, of which there are many others still awaiting explanation. The enquiry into the reason of the frambesial eruption may well produce a practical result, for if it were possible to prevent syphilis taking this form, we should thereby materially limit its infectiveness, and so lessen the amount of syphilis in the world. Here, then, is the field for research into yaws, which may yield a vast store of health for the unhappy natives of tropical lands.

Syphilis as yaws is almost general in tropical countries wherever the conditions of damp, heat and dense vegetation obtain. It is recognised that coko in Fiji, puru in the Malay Peninsula, parangi in Ceylon, galtoo in the West Coast of Africa, tomo in Samoa, are all identical with yaws in the West Indies. Yaws is well-enough known in every tropical country to bear a local name, and the synonyms are as numerous as the languages of the peoples affected. It was endemic in all parts of the tropical world before the European appeared on these scenes to confuse the geographical distribution of races and diseases by his facilities for inter-communication. Syphilis in all forms is now universal, and was probably so from very ancient times. Its wide spread proves, I think, that it must have been well established at an early period before the differentiation and distribution of human races.

It has been suggested by Hutchinson that yaws was the original disease prevalent in the tropics, and that it was introduced into Europe from them. Under the influence of a different climate, and the cleaner habits of the Europeans, it was limited and modified until it became usually venereal. He thinks that Sibbens and Button-scurvy were frambesial syphilis. Considering the wide

distribution of yaws, it is unlikely that the European countries could have escaped infection until modern times. The forefathers of the Caucasic peoples must have been already infected before they left the race-home on the pleistocene plateau of Sahara.

The experiments of Metchnikoff and Roux have shown that apes can be inoculated with syphilis. The femur of *Pithecanthropus erectus* shows evidences of disease very suggestive of syphilis. I am aware that these have been attributed to myositis ossificans, but one would think that the latter is more likely to be a recent disease resulting from the artificial circumstances of human life. In the Chaldean epic of Izduhar or Gilgames, it is told how the demi-god, having incurred the anger of Istar, was afflicted with a plague. An eruption of sores covered his body, his bones ached, his strength waned, his hair fell out. At length, under the advice of his beatified ancestor, Hasisadra, he was restored to health by sea-bathing. George Smith and other translators of the cuneiform script, following him, have read this to be leprosy. But every eruption on the skin is loosely so called by those writing on ancient matters. The leprosy of the Levitical regulations (Lev., chapters 13 and 14) was certainly not lepra. It no doubt included more than one disease, but most likely it referred chiefly to yaws. There is, perhaps, a reference to psoriasis in the " bright spots," and to frambesia in the appearance of the raw quick flesh. The disease was one of rapid development, for a suspect was kept under observation for seven days, and then for seven days more. Provision is made for restoring the outcast patient to society when he became clean. This would have been useless in the case of leprosy. The suggestion that Leviticus refers to yaws is a very old one, and has been discussed unfavourably by Alford Nicholls. But there is no common disease except syphilis which would explain all the regulations. True leprosy, no doubt, came under the operation of the laws, and was dealt with along with the syphilis.

It may be claimed that the symptoms of syphilis were sufficiently well known to find a place in the folklore of the early Semitic tribes, and to be subsequently included by the compilers of the traditional history and the authors of the great poems; and it was found necessary to devise an elaborate legislation to prevent its spread. The epic of Gilgames cannot be dated earlier than the second

millennium B.C. It has already enveloped in myth an invasion of the Elamites, which occurred about 2300 B.C. But even that is a respectable antiquity, and if syphilis was known to the poet of this the oldest epic in existence, there has been ample time and opportunity for it to spread to Europe before the middle of the second millennium A.D.

It is now known that endemic non-venereal syphilis may flourish even in Europe. Metchnikoff[1] points out that in rural districts in Russia the children are the chief sufferers and agents of the spread of syphilis, just as they are of yaws in the tropics. There is no reason for postponing the introduction of the disease into Europe to the 16th century, when it is far more probable that it has been present and spreading in a non-venereal manner as long as the human race has been in existence.

I append a few cases of yaws, that is, cases that displayed the frambesial eruption alone or, as is more usual, accompanied by other syphilides.

CASE 26.—C. W.; 8 years; Black.

January 23rd, 1906. Seen with an ulcer at the web of toes.

February 10th, 1906. Complained of pains, fever, and malaise, given liq. hydrag. perch.

March 24th, 1906. Onychia of nearly all the toes, whole body spotted with small round patches of scaly psoriasis. A large frambesia at the arms, which is said to have appeared a week before. Large glands in both groins. Ulcer on the plantar surface of web of toes is still in the same state as when first seen. Took six weekly injections of salalembroth. The frambesia disappeared rapidly, the skin cleared and the foot healed.

Discharged from treatment with directions to return in a month. Has not been seen since.

CASE 27.—C. B.; 3 years; Black.

March 1st, 1906. Has had yaws lately. One small frambesia still apparent at the angle of mouth. Had glands on elbows, in back of neck, axillae, below both angles of jaw, and in groins. Profuse eruption of small raw eroded surfaces, not raised but covered with moist yellow pellicles, in the left axilla. Few similar, drier and more crusted in the right axilla. Two such on the abdomen. Papules of similar character, some moist and some crusted, on and behind both auricles.

CASE 28.—M. P.; 8 years; Black.

March 7th, 1903. Had a sore on right ankle some months before. Large gland in right groin. Small hard glands on both elbows and many in the neck. Complains of pain in knees and ankles. Several marks of previous yaws on the legs. Several frambesiae with crusts on the right knee and the back. Papular eruption (lichen) between shoulder blades and round the axillae.

1. Harben Lectures, 1906.

CASE 29.—M. J.; 12 years; Black.

January 16th, 1903. Numerous small, perfectly-smooth limpets, which when removed are horny cups covering a granular raised papilloma identical with frambesia. These are over the face, hands, and body. They are about 6 to 10 mm. in size. A few of the papillomata have yellow crusts as is usual for frambesiae. On the left ankle is a spreading patch of a mildly lupoid appearance, the edge being active, and in the centre of this is an ordinary frambesia. This is probably the mother yaw. Treated with liq. hydrarg. perch.

February 19th, 1903. The limpets and horns are larger.

February 12th, 1904. A small rupia over left eyebrow, small tubboes on both soles. Circular wall of rupial scab on both hands at the base of the fourth and fifth fingers. Rhagades between the fingers of the left hand. No marks of the previous limpet eruption. This exemplifies a dry type of papilloma, and also shows later secondaries of rupial type.

CASE 30.—F. C.; age not noted; Black.

December 30th, 1902. Has a dry crusted frambesia on the wrist. Suppurative dermatitis of both palms and both soles. Multiple small pigmented marks as of a vesicular or pustular eruption on both legs. Psoriasis on both elbows and knees. Took pot. iod. for five weeks and left off attending.

I have seen papular groups, without scaliness, on the knees and elbows in syphilitic cases so often, that I doubt the opinion that psoriasis in this situation is never syphilitic.

September 14th, 1903. Tubboes on soles, and plantar and palmar psoriasis (crab yaws), keratitis. Voice raucous. This illustrates the polymorphism of the eruptions and the symmetry so characteristic of syphilis.

CASE 31.—C. B.; 30 years; White.

January 2nd, 1906. Has a child with yaws. Ulcer on heel about 20 mm. wide, with thickened raised edges, and ichorous discharge. This began as a pimple two months before. The first eruption was that now on the same foot and lower leg, which is a moist dermatitis. There are crusted ecthymata and aciniform eruptions all over the body and sparsely on the limbs and face. Slight fever every evening. Urine high coloured. Rheumatoid pains in shoulders. Paronychia of one finger. No enlarged glands. Given liq. hydrag. perch.

February 13th, 1906. Profuse small spots of silvery scaly psoriasis on both arms and thighs. In the bend of right elbow are three drying frambesiae. Several frambesiae round the trunk, and a profuse eruption of them on the left foot. Few on the right leg. Groin glands enlarged. No fever pains or malaise. All frambesiae are small and not much raised. Received a course of seven injections of salalembroth during which the smaller eruptions faded, but frambesiae continued to appear. Was rested for a month from mercury. At the end of this time the skin was nearly clean and only a few frambesiae left. Continued on liq. hydrag. perch.

CASE 32.—M. P.; 8 years; Black.

1906. Sent to hospital as a case of syphilis by a medical officer who had just come out. Frambesiae on two toes. Condyloma of anus. Condylomata of prepuce and circinate plaques on the glans penis. Enlarged groin glands. Circumcised and treated with six injections of salalembroth. Quite clean, and discharged.

CASE 33.—A. W.; 9 years; Black.

1906. Two large frambesiae by the side of a scabbed sore on the back of the left foot; and whole body, limbs and face covered with spots of psoriasis, even the palms of the hands. Hard glands in both groins, left being especially large. Glands on elbow.

CASE 34.—Female; 7 years; Black.

1906. Large irregular branched ulcer on a thickened base and with raised edges, on the back of the left heel. Glands in both groins and shotty glands in neck.

Several frambesiae on cheek and neck. Very large frambesiae at side and back of one knee, dry crusted papillomata on forehead and back of neck. These are essentially the same as yaws, but are dry, and have dark scabs. A symmetrical lichen covers both buttocks and back of thighs.

CASE 35.—L. K.; 19 years; Coloured.

April, 1906.—Had a venereal chancre last year accompanied by fever and rheumatoid pains. Glands in both groins enlarged. Profuse eruptions of frambesiform syphilides over both knees, extending to front of thighs and legs. These are about 12 mm. each. Some are moist with yellow pellicles, under which is a granular surface of elongated papillae. Some have dry brown crusts, which when removed expose a frambesial surface.

There are pigmented pits of a recent vesicular eruption on the right arm and the thighs.

The eruption in this case would have been called yaws by anyone. It was atypical in two respects. It was quite symmetrical, and the frambesiae were crowded together so as to be sometimes confluent. But neither of these features are exclusive of yaws. Such cases of yaws following a venereal infection are, perhaps, more common than is known. A syphilitic history is rarely enquired into when a patient displays yaws.

CASE 36.—M. W.; 6 years, Black.

July 16th, 1906. Oval, flat, callous ulcer, with a glazed surface and pigmented areola, on the front of the left shin. Enlarged glands in both groins and both elbows, several large glands in the neck.

A crop of small frambesiae with dirty-brown crusts on the forehead. Several larger flat condylomata with slight crusts, or naked with moist yellow surface, on the front of the neck and right cheek. Numerous papules varying in size scattered on the face. On the left cheek a group larger, and almost frambesiform. Small frambesia on the vulva. A profuse eruption of dry papules of all sizes up to 4 or 5 mm. over both buttocks and back of thighs. On the elbows a warty eruption of large papules with hard dry horny tops. Many of the papules elsewhere have a horny plug or horny apex of epithelium. There is a fine papular eruption on the front of the knees. Few scattered papules all over the skin. This case well exemplifies the polymorphism of the yaws eruptions. There were at least five syphilides of different appearance present at once.

TREATMENT OF SYPHILIS IN NEGROES

If we decide to treat yaws, whether it be syphilis or not, with mercury, it is necessary to appreciate that the treatment must be persevered in for many months. The frequent experience of the recurrence of frambesial eruptions after being presumably "cured" by mercury, indicates that the mercury must be continued, as in syphilis,

more or less through the first two years off and on. It will be found a very difficult thing to get a peasant to understand or act up to this, which in most cases will be only a counsel of perfection. When it comes to the tertiaries and potassium iodide, the patients readily learn from the relief obtained to value the drug. In St. Kitts and in Fiji the natives are fully alive to the uses of iodide, and prescribe it for themselves freely.

I have noticed doses as high as ninety minims of liq. hydrag. perch. mentioned in some of the literature on yaws, and there is very frequent reference to salivation. I wish to draw attention to the fact that negroes bear mercury badly. My father, a keen syphilist with many years' experience of treating negroes in hospital, where one can see best what one is doing, warned me of this. Fifteen minims three times a day is, I believe, about his limit. My own experience has amply borne out this. Though one is not afraid of "touching" the gums of a patient, and, in fact, generally likes to see them just "touched," as an indication that mercury is being pushed to its limit, yet salivation is undesirable. Apart from any physical effect it may have on the health or strength of the patient, it is apt to shake his confidence, and perhaps frighten him away altogether. It is astonishing how quickly some blacks do become salivated. I have seen a woman very badly ptyalised by five grains of calomel. Blue ointment applied to a wrist for a few days salivated a man, and a young woman became salivated before she had finished using half an ounce of blue ointment by daily inunction of about one dram. In the last case, there was certainly no idiosyncrasy; she had taken mercury, by the mouth in small doses, freely before, and has since received a course of intramuscular injection of 0·03 gme of salalembroth weekly, equivalent to about $\frac{2}{7}$ gr. of corrosive sublimate.

I have used mercury by inunction of blue ointment, mouth administration of liq. hydrarg. perch., calomel, and grey powder, and intramuscular injections of salalembroth. After a fairly large experience, I am confident that mercury is borne best when given by the last method. Inunction may salivate quickly and suddenly. In hospital one can watch it, but even so the ptyalism may occur suddenly. Grey powder with carbonate of iron is an excellent treatment, especially for children with hereditary taint or yaws. It

is convenient also for adults, for they can be given a hundred pills at a time, which last a month. I do not like to exceed 0·1 gm. thrice a day. Calomel I do not value for antisyphilitic treatment; but for general use in dispensary work the liq. hydrarg. perch. in a mixture is the most useful. I generally colour this lightly with methyl blue, which gives an impressive and distinctive value to the bottle. One should begin at the rate of 2 c.cm. of liq. hydrag. perch. per day—say, 10 minims three times a day, and increase to 3 c.cm. It is hardly necessary to go higher than this. Children bear this and any mercury well in proportion to the adults. For an infant of three months with inherited disease I give 0·15 c.cm. three times a day, equal to about two and a half minims.

Intramuscular injection of soluble salts is, however, far the best mode of administering mercury in my experience. I cannot speak of the insoluble preparations, for, being quite satisfied with the salalem-broth, I have not tried the emulsion of metallic mercury so highly spoken of by some. In spite of the pain, I have found that out-patients attend very well. Out of a number who were to take six injections each, the average attendance was five times. I have used this method on infants with hereditary syphilis also, and in yaws. My experience is not yet very large, but I have had abundant opportunity of proving that some negroes can carry up to 1·5 c.cm. of a 2 per cent. solution of mercury perchloride without any sign of spongy gums. This is equivalent to nearly half a grain of the salt, or about half an ounce of liq. hydrag. perch. Usually, I begin with 0·7 c.cm. and work up to 1·0, using a solution containing mercury perchloride 2 per cent., ammonium chloride 1 per cent, and giving one injection weekly. With care, washing the buttock with an antiseptic and then with ether, I have not yet seen an abscess result, and only rarely any induration. For hospital treatment, or for regular yaws dispensaries, the intramuscular method should be the most certain, the easiest and most effectual method of giving mercury. The treatment is shorter, and for that reason more useful in the case of natives. It should be possible to get most cases to attend for six or eight weeks for a course of injections, and repeat this twice more within the first year. After this, one attendance a month for another six months. The symptoms that may be expected should be explained to the patient, in order that he may at once report himself on the occurrence of any of them; and

certainly it is our duty to enlarge to any extent on the terrors of the tertiaries.

There is no necessity for special yaws asylums, but it is very desirable to have some provision for hospital treatment of severe and cachetic cases. Special wards in a general hospital would meet the requirements.

In the local treatment of yaws, I have obtained best results from a paint of zinc oxide and calamine, an old-fashioned but ever-useful preparation. It is readily applied by the patient or his parents; it is not easily wiped off, and, most important of all, it keeps the frambesia dry. I feel confident that a case of yaws is not very likely to infect other persons from his skin lesions as long as the frambesiae are kept dry and well coated with this paint. The risk of tonsillar infection from spoons and cups cannot, of course, be avoided in the peasant class, and that this is a frequent mode of infection with syphilis is well recognised.[1] But the most important material of infection in yaws is the secretion of the frambesia, and this can be limited by local applications, and by causing the absorption of the papilloma as rapidly as possible with mercury internally.

1. Kyle, Diseases of the Throat, p. 464.

A DESCRIPTION OF SOME GOLD COAST ENTOMOSTRACA

A DESCRIPTION OF SOME GOLD COAST ENTOMOSTRACA*

BY
W. M. GRAHAM, B.A., M.B.

It is a matter of common observation in the Gold Coast Colony, Ashanti and the Northern Territories that the incidence of Guinea worm varies locally. Some stations shew a high percentage of cases, some a low percentage, and some are free from the disease. Why should such local variations exist? The probable explanation is the abundance, the variety or the absence of the Cyclops-host from the water sources of the locality. Cyclops belongs to a widely-distributed family, and some species of the family can be found readily in the water sources of every station. But as Guinea worm disease is not present at every station, it is evident that the presence of any species in the water is not sufficient to cause the disease. The inference seems probable that a special species of Cyclops is required to act as efficient host to the worm, and that the absence of such species from the water sources of a station coincides with the absence of Guinea worm disease. If this be true, then an exact knowledge of the species to be found at each station becomes of great practical importance, but there is at present no classification of Gold Coast Copepoda. I have, therefore, sought in the present article to furnish means for the identification of the species found in the water sources of the station of Obuasi, and in those of the country within a three-mile radius round it, as a first instalment. Obuasi is a mining village on the Gold Coast Government Railway, 124 miles from Sekondi. The country is hilly and covered with forest. During the dry season the streams are small, sluggish and full of weed. With the setting in of the heavy rains they become rapid torrents, and the weed disappears. During the first period, to the end of April, Cyclops 1 to 5 can be caught in the streams, but after the first week in May

* The thanks of the Editors are herewith given to Dr. Brady for his kindness in identifying and describing this collection of Entomostraca.—EDS.

I have failed to find them there. The country is covered with gold holes of varying depth, and along the railway by borrow-pits. The water in the shallower pits drys up, and by the end of the dry season the bottoms of the pits have been baked dust dry. After a week's heavy rain these pits begin to retain a permanent pool of water. Upon this pool frog spawn soon appears, and shortly afterwards adult females of No. 6 (*Cyclops leuckarti*, Claus) can be found. By the middle of May the intermediate nauplius forms have appeared. Then the females become gradually less numerous and males increase in number, until by the middle of July the females have become rarer than the males.

Meanwhile, Cyclops Nos. 2, 3, 4 and 5 appear in the pools in relatively small numbers, and are soon accompanied by their nauplius forms.

Lastly, Cyclops No. 7 (*C. simillimus*, sp. nov., Graham) appears in the pools (the females first, the males later), and seems to take the place vacated by *C. leuckarti*.

This account brings up the history to the end of July; and I was transferred early in August to another station.

The inference previously alluded to, that all species of Copepoda cannot act as efficient hosts to Guinea worm, is strengthened by the following considerations:—

1st.—There is a large number of species.

2nd.—The habits of the different species vary greatly. Some are surface feeders, some are found at the bottom. Some inhabit foul, some clean water. Some leave the water to climb on stalks of water-weed enveloped in a drop of water carried with them, some do not leave the water. Some are found in streams, some are not.

3rd.—The different species are infested by different parasites. I have found Nos. 3, 4 and 5 infested only by ecto-parasites (algae), and No. 6 infested by ento-parasites (worms).

4th.—The different species differ in the date of their appearance in the pools. Some are found early in May; some appear, or, at least, only became numerous, in July. The importance of this variation in date has been already explained by me in a former article (B.M.J., 11/11/05).

A careful examination of the water sources of each station, and a classification of the Copepoda found in each place, is urgently required. When it has been made, a comparison of the species of Cyclops found at a station where Guinea worm is common with the species of Cyclops found at a station where Guinea worm disease is absent would, I have no doubt, offer an explanation of the observed local variation in the incidence of the disease. The practical value of an explanation is evident. It would enable the Medical Officer to identify those water sources likely to aid in the propagation of the malady.

CYCLOPS NO. 1. ? *Cyclops bicolor*, G. O. Sars.

MALE.—Colour yellowish, with darker coloured first antennae. First antennae, 11 segments as long as $\frac{3}{4}$ cephalothorax. Total length, 0·46 mm. Cephalothorax and thorax, 0·30 mm. Furca, 0·03 mm.

FEMALE.—Colour as in male. First antennae as in male, but more slender; total length, 0·62 mm. Cephalothorax and thorax, 0·42 mm. Furca, 0·04 mm. Egg sacs, a pinkish colour, carried apart.

In both series the outward tail sita is long. Found in pool with Calanus No. 1; water clear, and used by village as supply.

CYCLOPS NO. 2. *Cyclops varicoides*, sp. nov. (Brady)

MALE.—Colour, transparent pale yellow. First antennae, 12 segments, nearly as long as cephalothorax. Total length, 0·59 mm. Cephalothorax and thorax, 0·38 mm. Furca, 0·04 mm.

FEMALE.—Colour as in male. First antennae as above, but more slender. Total length, 0·64 mm. Cephalothorax and thorax, 0·39 mm. Furca, 0·04 mm. Egg sacs, a yellow colour, carried close together.

Found in river water.

CYCLOPS NO. 3. *Cyclops longistylis*, sp. nov. (Brady)

MALE.—Colour, a pale yellowish-green. First antennae, 12 segments as long as cephalothorax and two thoracic segments. Total length, 0·70 mm. Cephalothorax and thorax, 0·45 mm. Furca, 0·10 mm.

FEMALE.—Colour as above. First antennae as above, but slender. Total length, 0·84 mm. Cephalothorax and thorax, 0·50 mm. Furca, 0·12 mm. Egg sacs, a violet colour, carried slightly apart.

This species is frequently covered partially or entirely by ecto-parasites (algae).

It can be readily distinguished from No. 2 by the great length of the furca.

CYCLOPS NO. 4. *Cyclops virescens*, sp. nov. (Brady)

MALE.—Colour, cephalothorax a pale yellow-green; thorax a dark green. First antennae, 10 segments; copper coloured, carried a sparkling spot below the eye when swimming; an air bubble in mouth. Total length, 0·47 mm. Cephalothorax and thorax, 0·29 mm. Furca, 0·03 mm.

FEMALE.—Colour as above. First antennae as above, but lighter in colour. Total length, 0·60 mm. Cephalothorax and thorax, 0·40 mm. Furca, 0·04 mm. Egg sacs, pale yellowish, carried very close together.

Caught in rapid streams, and later on in year in ponds.

CYCLOPS NO. 5. *Cyclops pheleratus*, Koch

MALE.—Colour, a bright copper. First antennae, 10 segments, as long as the cephalothorax only. Total length, 0·60 mm. Cephalothorax and thorax, 0·38 mm. Furca, 0·05 mm. Legs of a light blue colour.

FEMALE.—Colour as above. First antennae as above. Total length, 0·77 mm. Cephalothorax and thorax, 0·46 mm. Furca, 0.08 mm. Egg sacs, a bright blue colour, carried close together.

This species leaves the water readily, and climbs on the sides of the vessel carrying a drop of water with it.

CYCLOPS NO. 6. *Cyclops leuckarti*, Claus

MALE.—Colour, a very pale yellow-green. First antennae, 17 segments, as long as the cephalothorax and thorax. Total length, 0·89 mm. Cephalothorax and thorax, 0·54 mm. Furca, 0·06 mm.

Become plentiful at end of July. I have not found males before the middle of July.

FEMALE.—Colour as above, but not so very pale. First antennae as above. Total length, 1·04 mm. Cephalothorax and thorax, 0·65 mm. Furca, 0·10 mm. Egg sacs (white) transparent, a long oval, egg very circular, sacs carried at an angle of more than 45°.

Found in stagnant pools in May. Become scarce in July.

CYCLOPS NO. 7. *Cyclops simillimus*, sp. nov. (Graham)

MALE.—Colour, very pale salmon with orange spots (circular) in cephalothorax. First antennae, 17 segments, somewhat longer than cephalothorax and thorax. Total length, 0·62 mm. Cephalothorax and thorax, 0·35 mm. Furca, 0·05 mm.

FEMALE.—Colour, pale yellow-green with circular orange spots in cephalothorax. First antennae as above, a very pale yellow. Length as above. Total length, 0·80 mm. Cephalothorax and thorax, 0·49 mm. Furca, 0·05 mm. Egg sacs, a pale yellow, carried apart.

This species also usually carries an air bubble in mouth when swimming. It differs from No. 6, being smaller and having relatively shorter tail setae.

CYCLOPS NO. 8. (*non det.*)

MALE.—Not found.

FEMALE.—Colour transparent, with slightly milky spots in cephalothorax and thorax. First antennae, 17 segments, as long as cephalothorax and thorax. Total length, 0·79 mm. Cephalothorax and thorax, 0·48 mm. Furca, 0·09 mm. Egg sacs small, milky white, carried apart.

Caught in well half a mile from the sea. Water in well clear, but slightly brackish. Frogs in well.

CALANUS NO. 1. = *Diaptomus innominatus*, sp. nov. (Brady)

MALE.—Colour, a very pale blue, the antennae being somewhat darker. First antennae, left 25 segments, right 22 segments, modified as clasper. Second antennae, 8 segments. Total length, 0·95 mm. Cephalothorax and thorax, 0·67 mm. Furca, 0·05 mm. Legs, central three pair a violet colour; fifth pair modified as claspers.

FEMALE.—Colour as above. First antennae, 25 segments. Second antennae, as above in male. Total length, 1·10 mm.

Cephalothorax and thorax, 0·75 mm. Furca, 0·07 mm. Egg sac, slightly blue, carried beneath.

When swimming appear transparent, with a dark blue longitudinal spot in about centre of cephalothorax and thorax.

I have only been able to find it in a single pond under trees, in which Cyclops No. 1 was also present.

CANTHOCAMPTUS NO. 1. = *Attheyella africana*, sp. nov. (Brady)

FEMALE.—Colour, a bright orange. First antennae, eight segments. Second antennae, four segments, forked. Total length, 0·35 mm. Cephalothorax and thorax, 0·17 mm. Egg sac nearly as long as body.

Very common in certain pools, where they occur in enormous numbers.

NOTES ON DR. GRAHAM'S COLLECTION OF CYCLOPIDAE FROM THE AFRICAN GOLD COAST

BY

G. STEWARDSON BRADY, M.D., LL.D., D.Sc., F.R.S.

(*Received May 14th, 1907*)

The following notes are intended merely as guides to the identification of species, and do not attempt a complete morphological account of the various forms. The brief descriptions are based upon the examination of females only, the males having been left out of the account.

In addition to the various species of *Cyclops*, the collection contained specimens of a minute Cyprid, a species of *Diaptomus*, and one called by Dr. Graham *Canthocamptus*, but which belongs to the nearly-related genus *Attheyella*. Both these last-named forms appear to be new, and I propose for them the names *Diaptomus innominatus* and *Attheyella africana*; they, however, need complete figures of structural detail, which at present I am unable to give.

GENUS CYCLOPS

No. 1. ? *Cyclops bicolor*, G. O. Sars.

No. 2. *Cyclops varicoides*, sp. nov.

Anterior antennae twelve-jointed, reaching to the third cephalothoracic segment, both branches of the first four pairs of swimming feet three-jointed, fifth pair rudimentary; caudal stylets short, about as long the last caudal segment. Length of body, exclusive of tail setae, 0·77 mm.

No. 3. *Cyclops longistylis*, sp. nov.

Anterior antennae twelve-jointed, reaching to the posterior border of the first cephalothoracic segment, both branches of the first four pairs of swimming feet three-jointed, fifth pair consisting of a

single small papilliform joint with two terminal setae; caudal stylets long and slender, about six times as long as broad, and equal in length to the three preceding caudal segments. Length of body, 0·78 mm.

No. 4. *Cyclops virescens*, sp. nov.

Anterior antennae ten-jointed, reaching to the third cephalothoracic segment, first four pairs of feet having both branches three-jointed, fifth pair minute, papilliform, bearing two long apical setae; caudal stylets short, about equal in length to the last caudal segment. Length of body, 0·6 mm.

This species is very similar to *C. gracilis*, Lilljebon, but differs in having all the branches of the swimming feet triarticulate; all the inner branches in *C. gracilis* being biarticulate; there are also other minor differences.

No. 5. *Cyclops phaleratus*, Koch

No. 6. *Cyclops leuckarti*, Claus.

No. 7. *Cyclops simillimus*, sp. nov.

Anterior antennae seventeen-jointed, reaching nearly to the posterior extremity of the cephalothorax, all branches of the swimming feet triarticulate, fifth pair biarticulate, last joint simple, narrow and bearing two long apical setae; caudal stylets about twice as long as broad, and nearly twice as long as the last caudal segment. Length, 0·77 mm.

EXPLANATION OF PLATE XXXIII

CYCLOPS BICOLOR, G. O. Sars

Fig. 1.—Female with ovisacs. × 74.
Fig. 2.—Male. × 74.

CYCLOPS VARICOIDES, n. sp., Brady

Fig. 3.—Female with ovisacs. × 74.
Fig. 4.—Male. × 74.

CYCLOPS LONGISTYLIS, n. sp., Brady

Fig. 5.—Female with ovisac. × 74.
Fig. 6.—Male. × 74.

The figures on this and the subsequent plates are reduced from Dr. Graham's original drawings.—EDS.

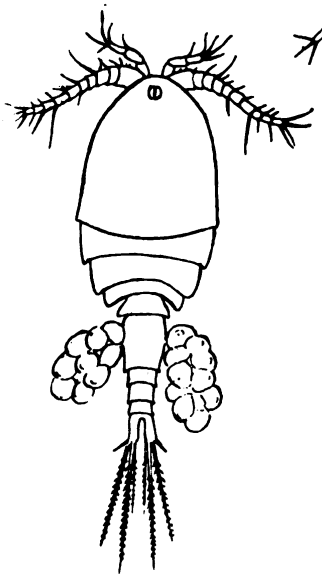


Fig 1

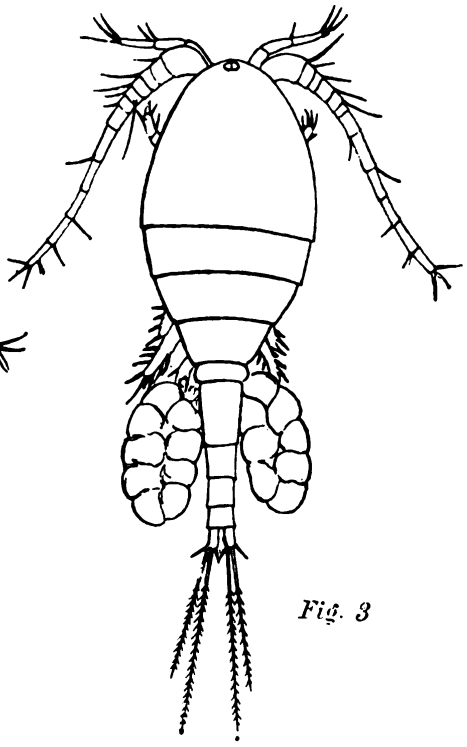


Fig. 3

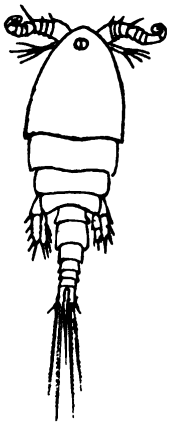


Fig. 2

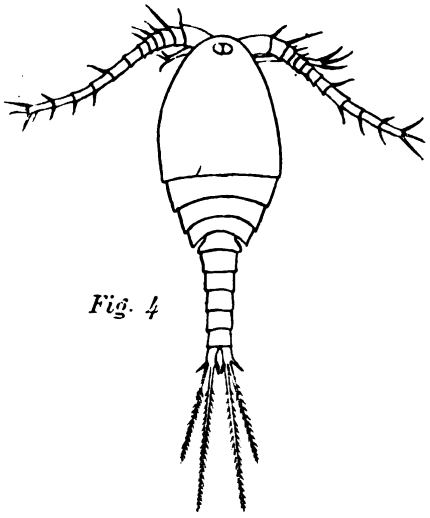


Fig. 4

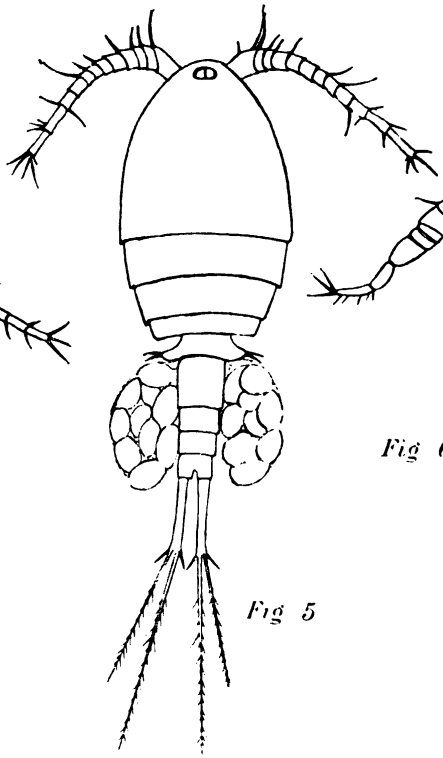


Fig 5

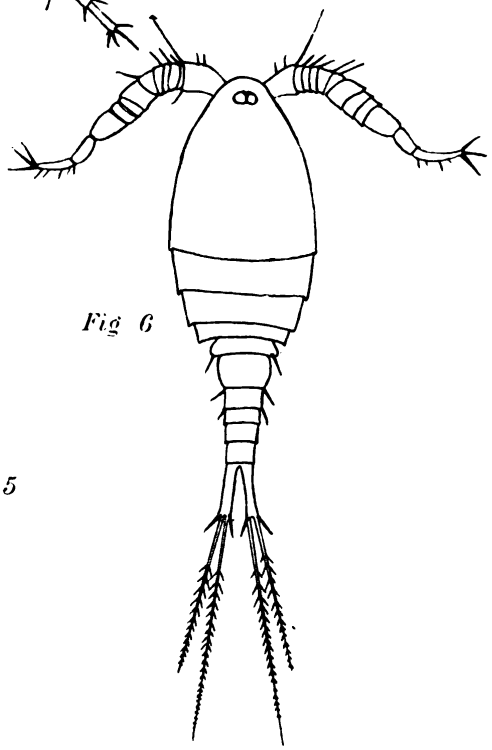


Fig 6

W. M. Graham, ad. nat. del.

EXPLANATION OF PLATE XXXIV

CYCLOPS VIRESCENS, sp. nov., Brady

Fig. 7.—Male. × 74.
Fig. 8. Female with ovisacs. × 74.

CYCLOPS PHALERATUS, Koch

Fig. 9.—Female with ovisacs. × 74.
Fig. 10.—Immature. × 74.
Fig. 11.—Male. × 74.

CYCLOPS LEUCKARTI, Claus

Fig. 12.—Male. × 54.

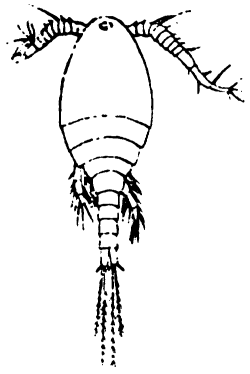


Fig. 7

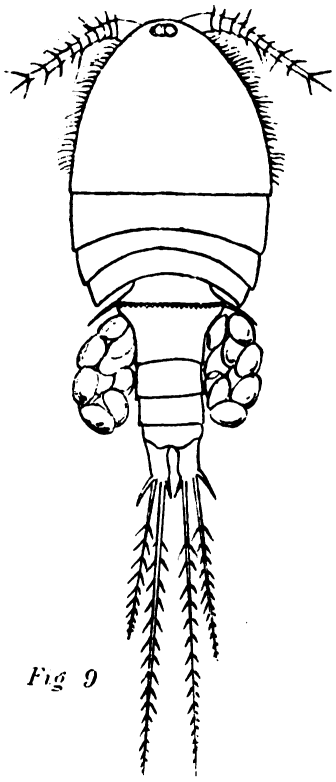


Fig. 9

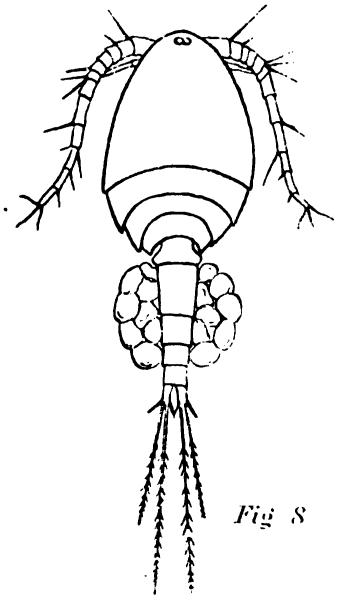


Fig. 8

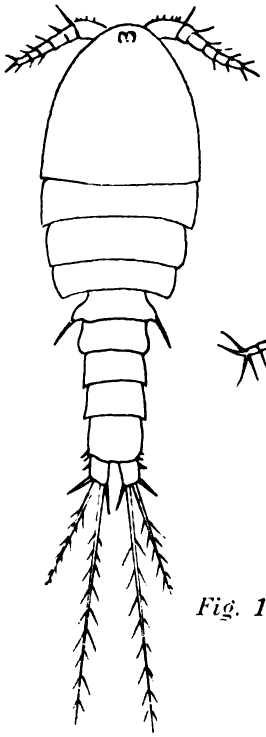


Fig. 10

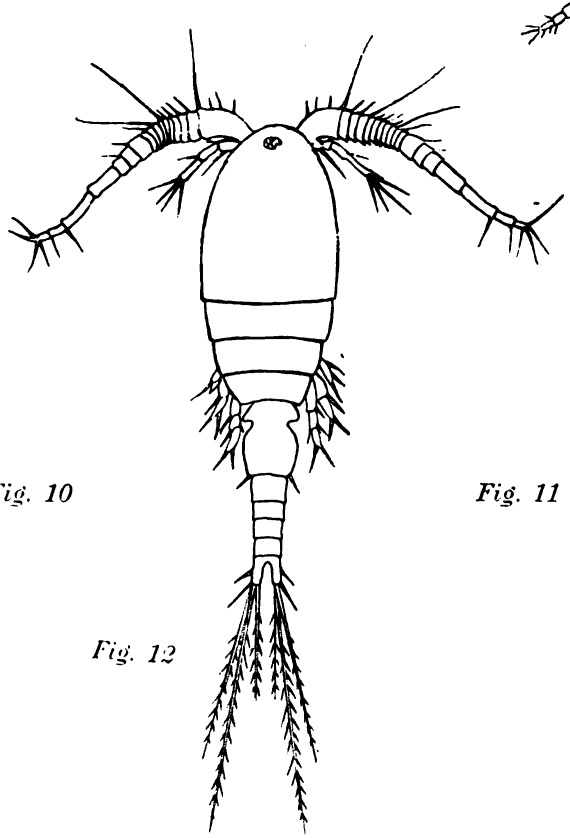


Fig. 12

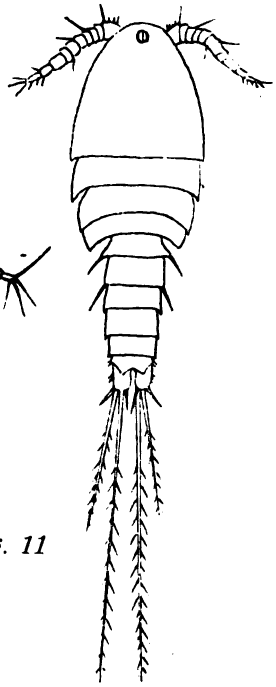


Fig. 11

W. M. Graham, ad. nat. del.

BB

EXPLANATION OF PLATE XXXV

CYCLOPS LEUCKARTI, Claus

Fig. 13.—Female with ovisacs. × 74.
Fig. 14.—Immature. × 54.

CYCLOPS SIMILLIMUS, sp. nov., Brady

Fig. 15.—Male. × 74.
Fig. 16.—Female with ovisacs. × 74.

CYCLOPS, ? sp.

Fig. 17.—There were no specimens of this species in Dr. Graham's collection when submitted to Dr. Brady for determination.

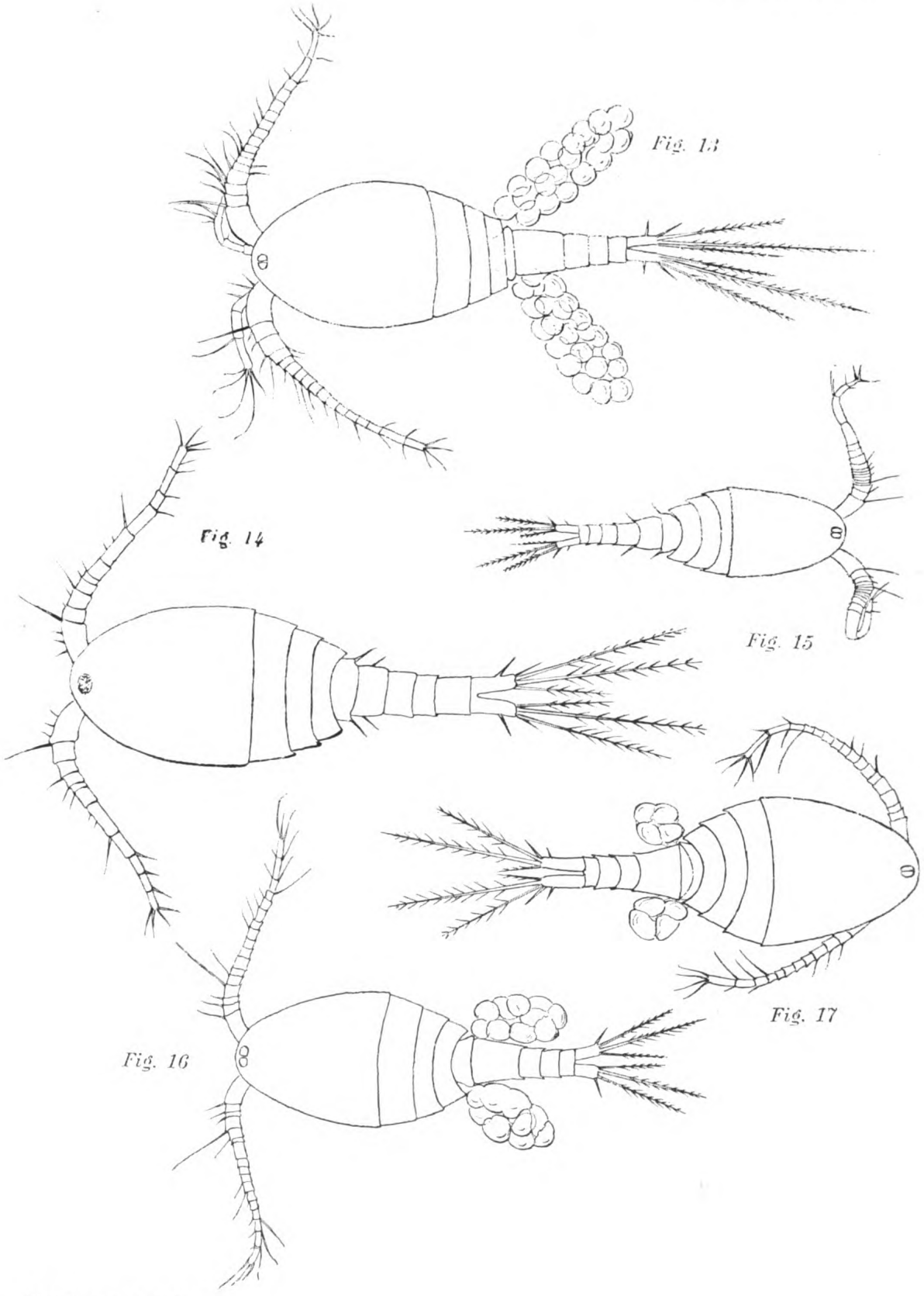


Fig. 13

Fig. 14

Fig. 15

Fig. 16

Fig. 17

W. M. Graham, ad. nat. del.

EXPLANATION OF PLATE XXXVI

DIAPTOMUS INNOMINATUS, sp. nov., Brady

Fig. 18. -Female. × 124.
Fig. 19. -Male. × 124.
Fig. 20. —Male, fifth pair of legs. × 102.
Fig. 21. -Female, fifth pair of legs. × 102.

ATTHEYELLA AFRICANA, sp. nov., Brady

Fig. 22. -Male. × 74.
Fig. 23. -Female. × 74.

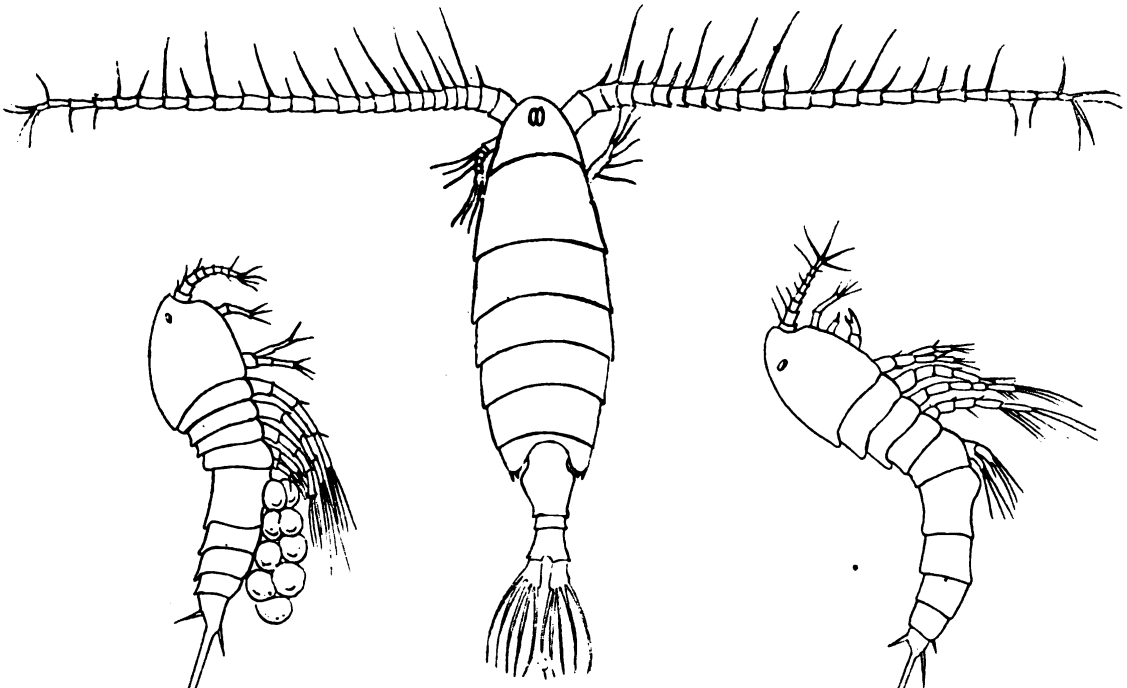


Fig. 18

Fig. 22

Fig. 23

Fig. 19

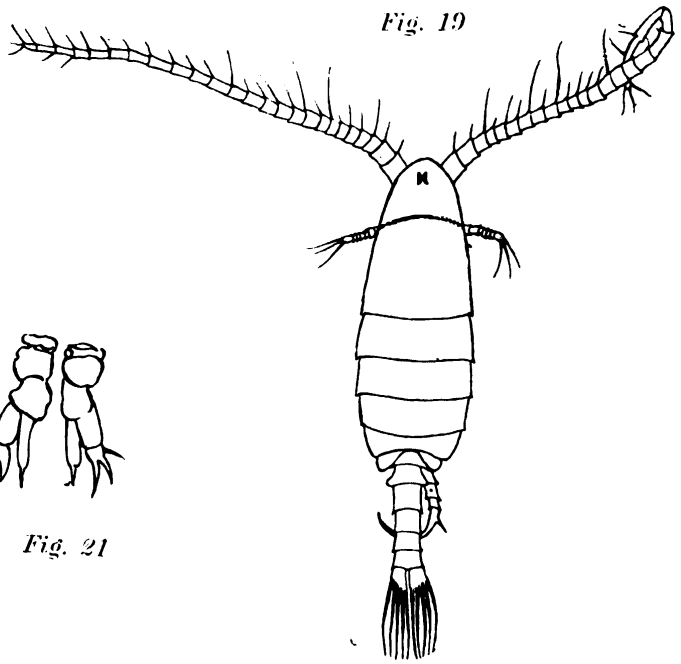


Fig. 21



Fig. 20

W. M. Graham, ad. nat. del.

ON THE MORPHOLOGY AND LIFE HISTORY OF *SPIROCHAETA DUTTONI*

ON THE MORPHOLOGY AND LIFE HISTORY OF *SPIROCHAETA DUTTONI*

BY

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From the Runcorn Research Laboratories of the Liverpool School of Tropical Medicine

(*Received July 1st, 1907*)

In the course of our experimental study of the spirochaete of African Relapsing fever, *Spirochæta duttoni*,[1] numerous films were made of the blood, and of the organs, of animals in the different stages of the disease, with a view of ascertaining the life-history of the parasite. Considerable uncertainty still exists concerning the morphology and the life cycle of the whole group of spirochaetes. While Novy and Knapp[2] deny that the parasites undergo any morphological changes, Prowazek,[3] in his work on fowl spirochaetes, gives them an undulating membrane and a definite nuclear apparatus, and also describes intra-cellular stages. One terminal flagellum was observed by Novy and Knapp in *Spirochæta obermeieri*, and by Stephens[4] in *Spirochæta duttoni*; Zettnow,[5] on the other hand, describes peritrichal flagella in *Spirochæta duttoni*, and Borrel[6] in fowl spirochaetes.

The stain employed in the present work was Giemsa's modification of Romanowsky's stain, in dry films. Wet films were not found to possess any particular advantage when working with this parasite.

The spirochaete consists of a darkly stained central core, which is surrounded by a light-stained periplastic sheath. This sheath extends beyond the central chromatic core, and is drawn out at one end into an elongated filament, thus forming a structure which has been described by various observers as a terminal flagellum. All attempts

to demonstrate peritrichal flagella, either in fresh or in stained specimens, have completely failed. The central core, or chromatic part, does not always stain uniformly, but in certain parasites lighter and darker areas are noticeable, either throughout the whole length or confined to one part of the parasite (figs. 1, 2). Very frequently, especially in spirochaetes which are disappearing from the circulation, the whole chromatic core seems to be broken up into an irregular number of granules (fig. 3). In this stage of infection the spirochaetes often show one or more swellings, either in the centre or at one end (figs. 4, 5). A fairly constant appearance, which has been previously described by various observers in *Spirochæta obermeieri* and *Spirochæta duttoni*, is a small unstained transverse band situated at about one-third of the length of the parasite (fig. 6).

A considerable amount of work was done with the object of demonstrating an undulating membrane. Although in specimens stained with Giemsa's solution an appearance was sometimes seen which resembled an undulating membrane, this was, in my opinion, due only to the flattening out of the spirals of the parasite. In wet films, even after a prolonged staining by Heidenhain's method, no trace of an undulating membrane could be seen.

The division of *Spirochæta duttoni* is, as a rule, transverse. The parasites increase in length and become thinner in the middle; this thinner part then elongates more and more until the two individuals separate (figs. 7, 8). It is very probable that the unstained area frequently seen in the normal parasite (fig. 6) is the point of the future elongation and subsequent transverse division. Occasionally, longitudinal division was seen to take place, especially at the time of the disappearance of the parasites from the peripheral circulation, and in this stage of the infection in the organs the parasite was seen to increase in thickness, the division commencing at one end of the spirochaete and gradually extending along its entire length (fig. 9). Judging from the scantiness of the parasites at this stage, it would appear that this process is a comparatively rapid one. In rare instances, at this stage of the disease, parasites were seen being engulfed by phagocytes.

A striking appearance, as depicted in fig. 10, was, on rare occasions, seen in the blood. Two spirochaetes were observed lying close to each other, touching at certain points. The one was stained

dark red with Giemsa, the other light blue, with apparently no chromatic core, but showing an irregular number of dark red granules situated at the points at which the two spirochaetes were in apposition. We are inclined to explain this appearance as conjugation.

Prowazek describes intra-cellular stages of *Spirochæta gallinarum* in the red blood cells. We were able to observe the same phenomenon in rare instances with *Spirochæta duttoni* just before the crisis set in (fig. 11).

Although the appearance of the parasites in the peripheral blood seemed fairly uniform, striking changes were observed in parasites seen in the organs, notably in the spleen, bone marrow, and liver.

Numerous spirochaetes, especially just before the crisis, when the blood was still swarming with parasites (principally in the spleen and bone marrow, rarely in the liver), were seen coiling themselves up (fig. 12), a few presenting a swollen appearance (fig. 13), the majority gradually becoming thinner and rolling themselves up into more and more complicated skein-like forms (figs. 14, 15) which seemed to become more irregular as the time of the crisis drew near. The majority of these forms were devoured by the phagocytes of the spleen, and at the time of the crisis the spleen cells were observed to be gorged with degenerated spirochaetes. In animals in which the spleen had been removed an analogous process took place in the liver.

A few similarly shaped parasites underwent a remarkable change:—The outline remained more regular for a time, and the parasite surrounded itself with a thin cyst wall, the interior of the cyst being filled with a faintly blue stained plasma (fig. 16). These forms, in scanty numbers, were to be seen even after all the other forms had disappeared. They apparently undergo further changes, as the shape of the parasite becomes more and more indistinct and, at a still later stage, only the cyst filled with small red granules persists.

We were unable to trace the further development of these forms, as in specimens stained by Giemsa's method it is impossible to differentiate them from blood platelets and other constituents. The fact that the filtrate of spirochaetal blood through a Berkfeld filter is infective suggests that these small granules may be the forms which pass through the filter and give rise to a fresh infection.

The life history of the spirochaete might be thus summarised:—Just before the crisis the spirochaetes disintegrate, certain of them coiling up into skeins, the majority of which are phagocytosed by the spleen. Some of them become encysted and break up into very small bodies, out of which the new generation of spirochaetes is evolved.

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~~Some forms described in the present paper were observed by this author~~.

EXPLANATION OF PLATE

The accompanying drawings were done with a Zeiss apochromatic objective 2 mm. aperture 1·4, ocular 18. Drawn to the scale of 4,500.

Figs. 1 to 12.—From the peripheral circulation of infected monkeys and rats, respectively.

Fig. 13.—From the liver of an infected monkey.

Figs. 14 to 16. Forms found in the spleen.

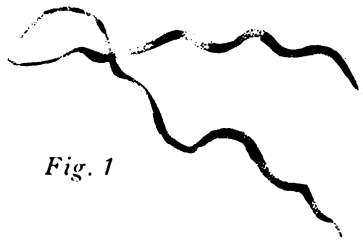


Fig. 1

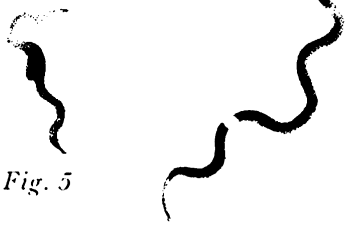


Fig. 6

Fig. 5



Fig. 2

Fig. 4



Fig. 3



Fig. 7

Fig. 9

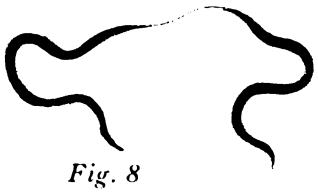


Fig. 8



Fig. 11



Fig. 10

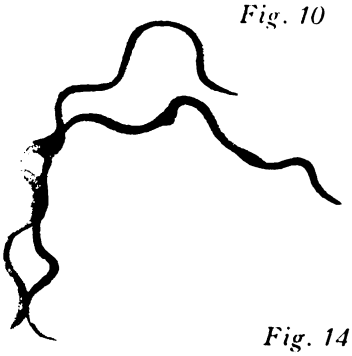


Fig. 14



Fig. 13



Fig. 15





Fig. 16

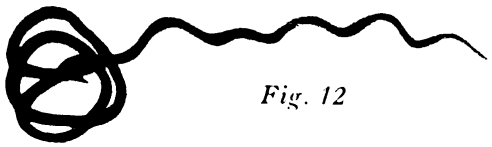


Fig. 12

PLATE XXXVII

THE
CYTOLOGY OF THE TRYPANOSOMES

THE CYTOLOGY OF THE TRYPANOSOMES

PART I*

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(Received July 15th, 1907)

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B. The morphology and the life cycle of *Trypanosoma gambiense*	448
C. The morphology, and the multiplication in the blood of *Trypanosoma brucei*	459
D. The morphology, and the multiplication in the blood of *Trypanosoma equinum*	459
E. Consideration of the foregoing observations	460
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(A) INTRODUCTORY

The trypanosomes belong to a group of organisms of great practical importance, since they are related to numerous diseases, not only affecting many valuable animals, but, in the case of sleeping sickness, man also. Notwithstanding the facts, the nature and

* A preliminary account of the observations relating to *T. gambiense* contained in the present paper was published in the *Lancet*, p. 1219, May 4, 1907. In a subsequent paper by Plimmer and Thomson received by the Royal Society, July 20, 1907, these authors appear to have encountered either the encysted Trypanosomes, or the resistant bodies (latent bodies) which we had previously described. But from the vagueness of their reference, Pro. Roy. Soc. B. Vol. 79, p. 509, it is impossible to be certain to which order of structures already described by us they do refer.

morphology of these organisms are as yet but little understood. Information upon these matters, as well as upon the various life cycles they appear to present, is greatly to be desired. Especially is this the case with regard to their morphology. The numerous descriptions of their structure and their metamorphoses already in existence have been drawn in general from the results of methods of research not calculated to produce any correct appreciation of their true cytological features. With very few exceptions, the study of the structure of the trypanosomes has been based, either upon what can be made out of the live animals, or else upon observations made upon material after it has been heated and dried, a method which, to say the least of it, may be shown, so far as the finer details of any cell structure are concerned, to be particularly barbarous.

Trypanosomes, like other unicellular organisms, can, however, be fixed in a great variety of ways which are commonly used during cytological research. The chief manipulative difficulty they present is the unreadiness with which they can be made to take any sort of differential stain. Still this difficulty is not insurmountable; and ordinary preparations may be produced which stain as completely as can be desired.

We have used the following fixatives:—Fleming's fluid; sublimate acetic acid; osmic acid vapour; osmic acetic acid vapour; and formalin vapour.

When the animals have been fixed, it is in all cases desirable to use somewhat special precautions in relation to the stain which may have to be employed, the process adopted depending upon the principle of applying a mordant, or mordants, before the actual stains are used. On the whole, we have found that the fixation with Fleming's fluid is unquestionably the best from a morphological point of view, while the staining methods through which we have obtained the sharpest colouration have been, on the one hand, the double safranine orange methylene blue stain invented by Breinl (see Appendix I); on the other, a slight modification of the Heidenhain iron haematoxylin process (see Appendix II).

As we are dealing in this Memoir (and in future publications which will be related to it) with many different specific forms, it is necessary in the first place to consider the cytology of the trypanosomes, as far as is now possible from a general point of view.

This course is unavoidable owing to the confused terminology which has grown up in the literature, and also on account of the present necessity of making clear the meaning we attach to different names. We have further to define our present conception of the nature of several structures which the trypanosomes possess.

When properly fixed, all the animals we have examined present an elongated cell form. No anterior or posterior extremity can, except in the most arbitrary sense, be defined. The exterior of the protoplasm is differentiated into a thin outer layer or ectosarc (periplast). Among the species with which this paper is concerned, the ectosarc is smooth, and does not present any definite ridges or stripes corresponding to the structure often described in the larger trypanosomes, such as those of the frog, and others.

The protoplasmic structure within the ectosarc consists of a very coarse *spongioplasm* (schaumplasma) containing fine staining granules embedded in its substance, the meshes of this spongioplasmic network being filled by a less stainable cytolymph. It is sometimes said that within the ectosarc, and distinct from the deeper portions of the spongioplasm, there exists a layer—the endoplasma. We have, however, not been able to demonstrate the existence of this sub-division.

The permanent cell structures contained within the ectosarc consist of a more or less central area, which, when subjected to Breinl's stain, assumes a purple colour (see figs. 4, 5, 11, 12, 13). We propose to call the whole of this area the *nucleus*. Within the nucleus there is always to be found a clearly-defined body which stains under the same conditions red, and we propose, for reasons which will become more apparent later, to term this body the *intra-nuclear centrosome* (Karyosome, Innenkorper).

It does not appear to be the case, when the animals are not dividing, that the nucleus can be correctly said to be bounded by any definite membrane. In most instances it appears more correct to say, that there is no definite membrane, but rather that there is a very sharp division between the spongioplasmic network and the finer network of the nucleus.

Near the broad end of the animal's body there is usually to be found a granule, or small group of granules, which stain like the intra-nuclear centrosome. These, whatever their numbers at any particular

period, we propose to call the *extra-nuclear centrosomes* (blepharoplasts). From one or more of these granules there springs a staining core, or flagellum, which lies in a thin expansion of the ectosarc, forming the so-called undulating membrane.

For present purposes we have thus the following terminology :—

Ectosarc = (Periplast).

Spongioplasm = (The substance of the network of the protoplasm).

Cytolymph = (The substance between the meshes of the spongioplasm).

Intra-nuclear centrosomes = (Karyosomes, Innenkorper).

Extra-nuclear centrosomes = (Blepharoplasts, micro-nuclei centrosomes, nucleoli).

Flagellum.

Undulating membranes.

In none of the trypanosomes which we have studied have we found the slightest indication of the existence of the so-called males, females, and indifferent forms. We have found that the often-asserted existence of these three types in the blood, a suggestion originating chiefly from Schaudinn,* is totally misleading.

So far as is at present known, trypanosomes are parasites inhabiting the blood, and body fluids, of a great variety of animals. Hitherto no non-parasitic forms have been discovered. As is the case with other parasites of this description, their life histories appear to have become modified to secure their transference from one host to another in different ways. When introduced into the blood and tissues of a suitable host, they usually multiply by fission until either the noxious influence of the infection destroys the host, as in the case of some strains of *T. gambiense* introduced into rats, or the infection runs a different type of course. In the latter type of infection the multiplication of the parasites in the blood rises to a first maximum, and then falls, so that the numbers may decrease to zero. After this fall, which we may speak of as the first negative phase, the parasites reappear, and reach a second maximum, and so on. In such cases, the infection follows an irregular course, which can be easily understood from the diagram given on page 449.

* For further information respecting this matter see Thomas and Breinl, Liverpool School of Tropical Medicine, Memoir XVI.

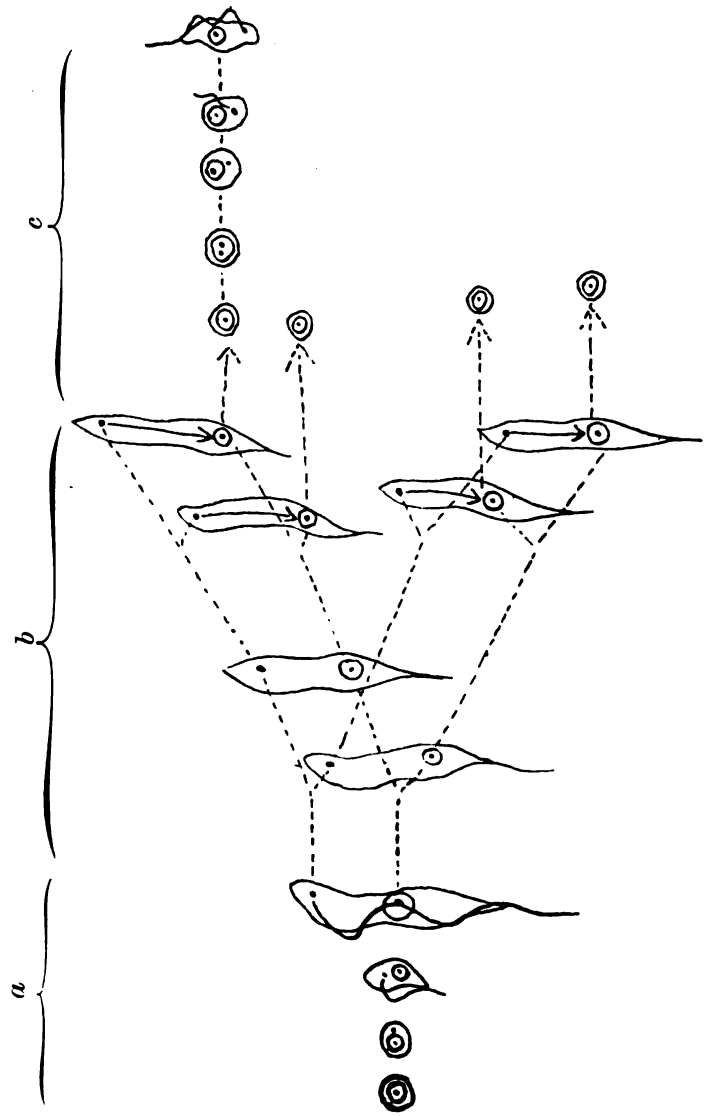


DIAGRAM.—Showing the life cycle of *Trypanosoma gambiense* in a rat:—

a. Development of the Trypanosome from the latent body.

b. The fission of the Trypanosome and the formation of the black line.

c. Reproduction of the latent bodies and the development of the Trypanosome therefrom.

It is a very remarkable fact that in some examples of the latter type of infection the blood during the negative phases, although apparently containing no trypanosomes at all, and even if it be properly filtered, is still capable of infecting other animals into which it may be introduced. These observations suggest the existence of another stage in the life history of the organism in the same host during such periods.

According to Schaudinn,* *Trypanosoma noctuæ* which infests the blood of owls in certain parts of Southern Europe is transmitted from owl to owl by mosquitoes. When withdrawn from the blood of the owl into the body of the mosquito the trypanosomes pass through a definite sexual stage, and are again reintroduced through successive bites, in another stage of their life cycle into the owls once more.

It is very desirable that these observations should be repeated. They have never been properly confirmed; but, assuming them to be correct, it by no means necessarily follows that a similar life cycle is pursued in the case of other trypanosomes with which we are now acquainted.

There are, indeed, numerous indications which we shall have to consider in more detail later, that render it probable that a life cycle such as that described by Schaudinn, involving successive hosts wherein sexual and asexual stages alternate, is the exception rather than the rule. Consider in this connection the following facts:— The trypanosomes *T. gambiense*, *T. brucei*, *T. equinum*, *T. equiperdum*, *T. lewisi* and others, can be transferred from animal to animal by simple inoculation of blood. Simple inoculation of blood can be effected by many sorts of flies. It appears to be so effected in the case of sleeping sickness through *Glossina palpalis*; but no sexual stage has hitherto been discovered in this fly. The observations of Bruce† showing that the infectivity of flies disappears after 48 hours, together with those of Dutton, Todd and Hanington,‡ who made a large number of experiments with regard to this matter on the Upper Congo, all seem to indicate that the infection by these flies is perhaps rather in

* Schaudinn. Generations und Wirtswechsel bei Trypanosoma und Spirochaete. Arb. a. d. kaiserl. Gesundheitsamte, Bd. XX, H. 3.

† Bruce, Nabarro & Greig. Reports of the Royal Society. Sleeping Sickness Commission, No. 4, 1903.

‡ Dutton, Todd and Hanington. Trypanosome transmission experiments. Ann. Trop. Med. and Parasit., Vol. I, No. 2, p. 199.

the nature of an accident than a necessary process involved in the normal life cycle of the parasites. Some such conclusion is borne out by other facts in relation to trypanosome infection. Thus in the case of *Dourine*,* simple inoculation of blood will transmit the disease, but it is habitually communicated amongst horses in quite a different manner, namely by coitus. Consequently, if there is a sexual stage in the life history of *Trypanosoma equiperdum*, this sexual stage must occur normally in the body of the horse. Further strains of trypanosomes, such as those of sleeping sickness and *Dourine*, may be kept for years in our laboratories through inoculation from animal to animal. In fact, such strains may be continued in this way for a quite indefinite period, a process involving an endless number of generations in the blood, and it consequently follows that if in such forms the sexual stage occurs only in some other host, this phase can be dispensed with for an altogether indefinite period.

As a matter of fact, there are yet other observations bearing upon Schaudinn's researches, which if they do not necessarily render his account of the sexual act improbable, seem to clearly indicate that it may exist in the instance of *Trypanosoma noctuæ* as a very unusual exception, an exception which may be incapable of throwing any general light upon the life history of the great group of organisms to which *Trypanosoma noctuæ* belongs. We may refer also particularly to the author's account of what he regards as the reduction process.

This, according to Schaudinn,† amounts to a sexual determination, or differentiation, accomplished through a nuclear division. That is to say, there occurs in *Trypanosoma noctuæ* a division (heteropolar mitosis, Schaudinn), which separates the female moiety of an hermaphrodite nucleus from the male. In other words, Schaudinn resuscitates (although he does not appear to allude to this fact) Balfour's and Minot's view of the formation of the polar bodies, and the extrusion of the so-called residual corpuscle during the formation of the spermatozoa.

* Rabinowitsch and Kempner. Centralblat für Bakt. Bd. XXXVII, H. 5.
See also Minchin, Gray and Tulloch. Pro. Roy. Soc., London. Vol. 78, 1906.
See further Laveran and Mesnil. Trypanosomes et trypanosomiases. Paris, 1904.

† Schaudinn: Neuere Forschungen über die Befruchtung bei Protozoen. Verhandlungen der deutschen zoologischen Gesellschaft auf der XV. Jahresversammlung. Leipzig, 1905.

It is necessary to be quite clear about this matter. The hypothesis respecting the function of the polar bodies, and so-called residual corpuscle of the spermatozoa, as the means by which the opposite "sex-stuff" is got rid of from the ovum, and the spermatozoon, has for various reasons collapsed some years ago. In the first place, the polar bodies cannot be homologised in any way with the residual vesicle. In the second place, it has been clearly demonstrated that in the vast majority of animals and plants the sexual reproductive cells, ova, and spermatozoa are precisely equivalent so far as their nuclei are concerned.* Reduction as now understood in animals and plants is not sex differentiation, but a process which results simply in the halving of the number of chromosomes in those cells which are destined to conjugate.

A great deal of confusion has been produced by Schaudinn's inaccurate use of the term reduction, a term which in general biology has long had a limited and a definite meaning. In dealing with these matters it must, therefore, be clearly understood that by reduction Schaudinn means sex differentiation, and that the term reduction in general biology does not mean sex differentiation, and stands for something quite different.

(B) THE MORPHOLOGY AND LIFE CYCLE OF TRYPANOSOMA GAMBIENSE

Trypanosoma gambiense (Dutton) is a parasite associated with the disease appearing in Equatorial Africa and known as Sleeping Sickness. It can be transferred by simple inoculation into nearly all the animals generally used for laboratory experiments. The infection may run a very varied course. Thus the strain used in Liverpool for inoculation into rats may simply increase until, within a period of a few days, the rat's blood is teeming with parasites and the animal dies from the effect of the invasion. In other cases among the rats, as in man, the number of parasites in the blood rises and falls in a

* It is unnecessary to refer to the vast literature of this subject. The repeatedly confirmed observations upon which the above statements are based, being so well known as to render it unintelligible why Schaudinn did not himself make it clear that the process he describes, under the term reduction, can have nothing in common with the reduction process as described and studied in animals and plants for the last 20 years. It is equally unintelligible why this author did not point out the identity between the process he discussed and that erroneously supposed to exist among animals and plants by Balfour and Minot.

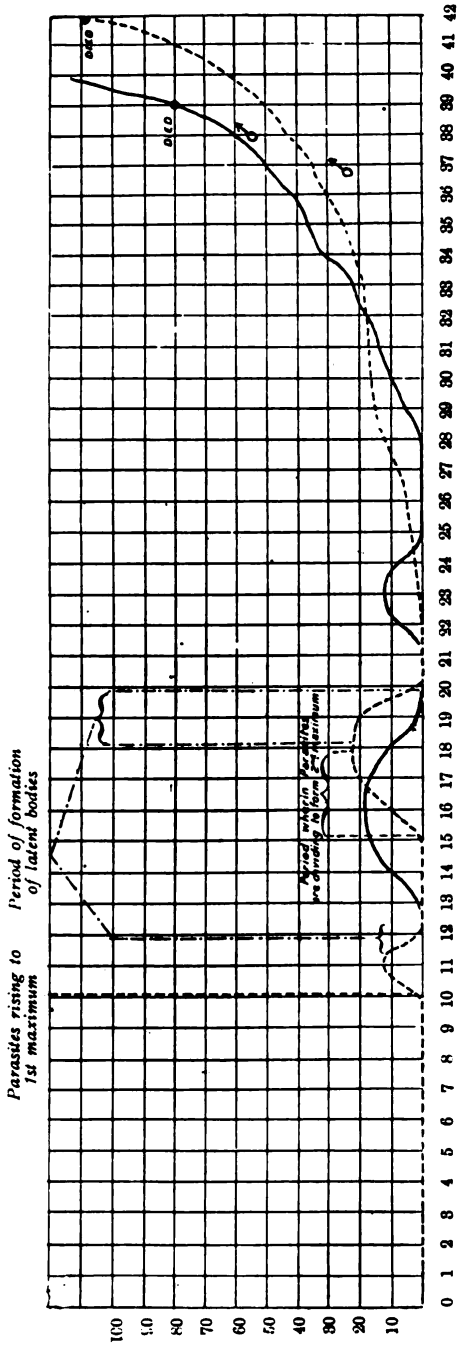


CHART OF TWO MALE RATS INOCULATED WITH *Trypanosoma gambiense*.

The horizontal figures represent the days after inoculation: the vertical figures the numbers of parasites in a microscopic field of blood; the curve representing the variation in this during the course of the infection. The two different curves represent two different infections.

somewhat irregular series of well-marked periods, the kind of oscillation produced being indicated in the chart given on page 449. When an animal has been previously infected, it has been found that even at a period when no parasites can be detected in the blood, the blood is nevertheless capable of infecting other animals by sub-inoculation.

As is already well known, *Trypanosoma gambiense* can be transmitted from animal to animal by the bites of flies; but the observations of Bruce* and others have shown that if more than 48 hours elapse after the flies have fed on an infected animal, subsequent bites produce no infection. The observations of Dutton, Todd and Haningtonf made on the Congo, show further that it is often extremely difficult to infect at all with flies, and the authors sum up the position in respect to this matter in the following paragraph:—

"We believe either, (I) That something is wrong in the way in "which *Glossina palpalis* has been used in these experiments, or, "(II) That *Trypanosoma gambiense* can be conveyed by some other "means than by it."

So far, then, from it being established that Sleeping Sickness is normally spread among the African population by the bites of *Glossina palpalis* alone, it would seem that the most recent work on this subject indicates that possibly the infection through flies is in the nature of an accident, and that the means by which Sleeping Sickness spreads, in the manner in which it does spread in the African interior, has yet to be discovered.

Trypanosoma gambiense as it appears in the Blood of an Infected Animal

When examined in the blood *Trypanosoma gambiense* is found to vary in size very largely. Thus from forms smaller than those represented in figs. 1, 2, 4, 6, we may select a series increasing gradually to the extreme dimensions represented in figs. 8, 9, 10.

From our own observations of the parasites as they appear in the blood, it does not seem possible to detect any true dimorphism, or trimorphism, corresponding to the so-called male, female, and

* Bruce, loc. cit.

† Dutton, Todd and Hanington, loc. cit.

indifferent forms described by Schaudinn,* Minchin,† and many other authors. The present examination of the forms appearing in the blood leads us to believe that there is to be found among these trypanosomes a series extending from those which are relatively small, to those which are relatively immense. The three forms often described and alluded to as distinct, consequently appear to be arbitrarily chosen examples in a continuous series of dimensions.

Multiplication in the Blood

From the time when the parasites appear in the blood of an infected animal until their numbers reach any particular maximum, rapid multiplication takes place by longitudinal fission of the individual trypanosomes, the multiplication being most rapid near the successive periods of maximum numbers. When the parasites are not dividing, they present the appearance represented in figs. 1 and 4.

The nucleus is nearly in the middle of the long axis of the cell, and consists of an outer stainable mass, enclosing generally a lighter central area, within which there lies a small, sharply-definable body, which stains red in contrast to the purple colour of the outer mass when subject to Breinl's stain. This central structure forms the intra-nuclear centrosome (karyosome). At the broad end of the cell there exists another staining granule which, when the cell is not dividing, remains single, the granule in question forming the extra-nuclear centrosome (blepharoplast, micro-nucleus, centrosome). It stains, under the above conditions, like the intra-nuclear centrosome. Arising directly from the extra-nuclear centrosome, there extends a delicate thread, which stains more faintly, but in the same manner as the centrosomes. It is enclosed in a thin expansion of the ectosarc running along the entire length of the cell. The thread projects at the narrow end of the cell as a long stained whip-lash. This thread forms the so-called flagellum, and the ectosarcal expansion the undulating membrane.

The first sign of an approaching fission is generally apparent in relation to the extra-nuclear centrosome. From this there buds out a small fragment, figs. 1, 2, and 3, which may become flattened, as in

* Loc. cit.

† Pro. Roy. Soc., 1907.

figs. 1 and 7, and can be seen to be attached to the original extra-nuclear centrosome by faint unstaining strands, fig. 7. At a later period there grows out from the new extra-nuclear centrosome a young flagellum, which gradually extends parallel with the old flagellum, as in figs. 2, 3, 6.

At a period of the fission roughly corresponding to the above, it can be observed that the intra-nuclear centrosome has also divided in the manner represented in figs. 2, 3, and that the staining outer portion of the nucleus has gradually collected around, and beyond, the dividing portions of the intra-nuclear centrosome. In this form of nuclear division characteristic of *Trypanosoma gambiense*, no chromosomes are formed at all, and the intra-nuclear centrosome behaves precisely as the so-called karyosome, or centrosome, during the division of the nucleus in *Euglena*, *Eimeria schubergi* (Schaudinn), and other protozoa. At a later stage the division of the intra-nuclear centrosome becomes complete, and the outer staining portion of the nucleus collects round the two intra-nuclear centrosomes as in figs. 3, 7, 8, forming together with these two bodies two daughter nuclei, having just the same appearance as the parent nucleus. For all practical purposes the nuclear division of *Trypanosoma gambiense* is thus amitotic; but it is a form of amitosis somewhat complicated by the presence of an intra-nuclear centrosome.*

In the remainder of the cell, the process of division proceeds by the extension and growth of the new flagellum through stages such as represented in figs. 11, 12, 13, 14, so that two apparently similar flagella are produced, each enclosed in an expansion of the ectosarc. They form separate undulating membranes and stretch from one end of the animal's body to the other, fig. 12. The extremities of the flagella become eventually separate, and the animal gradually splits from the narrow end towards the broad end, figs. 13, 14. The daughter forms finally separating, pass through stages such as those represented in figs. 13, 14. In this way we have eventually two trypanosomes, each exactly resembling the parent form, but smaller.

During the growth and division of the parasites in the blood, it is frequently possible to find large numbers of cells in all stages of

* In connection with this matter it should be noted that in many of the metazoa where the centrosomes are always extra-nuclear, these bodies may be definitely related to amitosis when this form of division occurs.

division, and rest, wherein there exist, especially towards the narrow end of the cell, quantities of granules that stain under the action of Breinl's stain an intense red. The colour of these granules is quite distinct from that of either the intra-nuclear centrosome or the extra-nuclear centrosome, and they cannot be stained with any satisfaction at all by means of the iron haematoxylin method. They are quite irregular in size and number, and also in their appearance, that is to say, they may appear in all the trypanosomes in all their stages at one period, and not at another. From these circumstances, we are inclined to regard these granules as of metabolic origin, and we can find no evidence that they arise from the nucleus at any time.

These bodies should, however, not be confounded with the minute granules always seen, more or less throughout the spongioplasmic network of the entire body. These latter may, and very often do, stain in the same manner as the intra- and extra-nuclear centrosomes, but we have been unable to find any indications as to their origin, or that they any more than the large metabolic granules have any special relationship with the nucleus.

It is possible, indeed probable, that some of these granules may correspond to the vegetative and trophic chromidia observed in *Rhizopods* by Schaudinn,* and by Hertwig† in *Actinosphærium eichhorni*, but owing to the very different methods we have used, we are not in a position to make any definite statement with respect to this matter.

At a late stage of division, such as that represented in figs. 14 and 13, the appearance of the organisms at first sight very much suggests an act of conjugation, but in all such cases that we have examined we have found no indication in relation to the nuclei, or the centrosomes which would suggest a conjugational act.

Along with the regular method of division just described certain modifications are frequently observed, which, although producing the most striking appearance (see figs. 8, 9, 10) are nevertheless apparently always capable of being explained through a disparity in the stage simultaneously reached by different parts of the cell. Thus the nucleus may divide completely and then divide again so as to form

* Schaudinn. Arbeiten aus dem kaiserlichen Gesundheitsamte, Vol. XIX, 1903.
† Hertwig. Archiv. für Protistenkunde, Vol. 1.
See also Mesnil. Chromidies et questions connexes. Bull. Past., Tome III, p. 313.

four nuclei, without, however, the extra-nuclear centrosome having divided more than once. Or the extra-nuclear centrosome may divide and form three or four flagella without the nucleus having divided at all. When these unusual methods are adopted, the gigantic trypanosomes figured on Plate I are frequently produced.

Changes in the Trypanosomes relative to the Stage of Infection

The appearances described in the preceding paragraphs are those which are encountered among the parasites during multiplication after their first appearance in the blood of the infected animal. If, however, other stages of the infection be studied there are to be found different morphological appearances among the parasites, which are apparently of the greatest importance. In the case of an animal which has become infected with *Trypanosoma gambiense*, and shows a well-marked oscillation in the course of the disease, it is found that as the number of the parasites is rising in the blood—that is to say, along the ascending portions of the curve in the diagram (page 449)—the phenomena presented are the same as those found in the initial stages of the infection which we have just described. If, however, we study preparations made at or near the maxima of the curve, other changes are found to be taking place in the trypanosomes besides those of simple division.

At the time the curve approaches any maximum, there are to be found trypanosomes which present the appearance shown in figs. 15 to 20. From this figure it will be seen that such trypanosomes are distinct from those which have been previously described, in that a relatively-thick stainable band proceeds from the extra-nuclear centrosome. This thick band is found to be most readily stainable by iron haematoxylin; it is less readily, but still stainable by the various aniline colours which we have employed. It grows from the extra-nuclear centrosome not along the surface of the animal as in the case of the new flagella, but down the interior of the cell towards the nucleus (see figs. 15, 16).* This stainable band, which appears near

* It is probable that the band we here refer to is the same as the structures described by Prowazek, Studien uber Saugethier trypanosomen. Arb. a. d. kaiserl. Gesundheitsamte, Band XXII, 1905, and also Miss Robinson, Notes on certain blood-inhabiting protozoa. Proceedings of the Royal Society, Ed. Vol. XVI, No. 6.

the periods of the greatest number of parasites in the blood, is fully twice as thick as the flagellum. As it increases in length it may reach, or even pass the nucleus; or it may become coiled upon itself, as in figs. 16, 17, 18. But whatever form it takes at first, the later appearance suggests that the band eventually becomes in one way or another definitely connected up with the nucleus. This suggestion is strengthened by the subsequent behaviour of the band, for it is seen eventually to become gradually less stainable, to break up into a string of fragments, and finally to disappear. Through all these later stages of its existence it is most certainly suggested, as in figs. 19, 20, that it is directly related to the nucleus, that is to say, to have been, or to be actually connected up with it.

We appear then, at or near the maximum number of the parasites in the blood, to have direct evidence of some sort of interaction taking place between the extra-nuclear centrosome and the nucleus. This phenomenon occurs only among animals in which no evidence of nuclear division, or cell division, is apparent. In such cases, we do not find that either the extra-nuclear centrosome or the nucleus is divided, and throughout the whole development of the stainable band the nucleus remains in a condition of complete repose.

If now we examine the portions of the curve of infection where the trypanosomes are decreasing in number, still other phenomena make their appearance. The numbers of the parasites gradually decrease in the peripheral blood, and at this time, in such organs as the lungs, the spleen, and the bone marrow, we find curious changes to be taking place in vast numbers of the trypanosomes encountered in these situations. Parasites showing the present changes are rarely found in the organs named above during the period when the number of the parasites is increasing in the blood, but at the time when the numbers are beginning to decrease we see in the lungs that numbers of trypanosomes show alterations in their nuclei. The protoplasm of the animal's body becomes detached from the periphery of the nucleus which lies in a clear space, while the nucleus itself contracts somewhat, and at the same time a large clear vesicle gradually grows up in connection with it, in the manner represented in figs. 22, 23. Round the outside of this vesicle and the nucleus there may be seen a layer of protoplasm enclosing in a delicate sheath both nucleus and vesicle together. When this stage has been reached, the rest of the cell body

rapidly disintegrates, the outline of the cell becomes lost, and the flagellum together with the extra-nuclear centrosome may be frequently seen detached, and lying loose among the cells of the organs examined (see fig. 26).

These phenomena, as we have said, are encountered in the lungs, but they are also found in the bone marrow and the spleen. After the above stage has been reached the nuclear bodies and the disintegrating remains of the parent cells disappear to a large extent from the lungs, the nuclear bodies being subsequently found in very large numbers both in the spleen and the bone marrow.

At first sight, the appearance we have just described might be supposed to be simply a phase in the disintegration of the parasites, and this was, as a matter of fact, the view which we were at first inclined to adopt. There are, however, reasons for thinking that, although the greater part of the protoplasm forming the bodies of the parasites undoubtedly does disappear, the peculiar nuclear structures we have just been considering do not follow the same course. After their detachment from the rest of the disintegrating cell body, the nuclear corpuscles become impacted in great numbers in the spleen and bone marrow. Here, instead of disappearing in the course of a few hours, they may remain intact for as much as ten days or more, in fact, throughout the whole of the negative or latent period of the infection, when no parasites are present in the peripheral blood. From the time of their formation, these peculiar bodies,* which we propose to term *latent bodies*, may be found in small numbers in the blood, and although they thus remain intact for a relatively long period, it

* The formation of these bodies appears not to have been previously observed at all in *Trypanosoma gambiense*; but similar bodies have been seen in relation to other trypanosomes, and have been variously interpreted. Thus, Rodet and Vallet, Arch. des Med. expériment., Vol. 18, No. 4, describe bodies which appear to be similar to the latent bodies of *Trypanosoma gambiense* in *Trypanosoma brucei* in the blood of the organs and regard them as degenerate forms. Plimmer and Bradford (Quar. Journ. Micro. Sci., Vol. 45) seem also to have found them in the spleen, &c., during *Nagana* infections, and describe them as apparently the nuclei of a plasmodial production. Laveran and Mesnil describe apparently similar bodies as "formes d'involution." Lingard (Journ. Trop. Vet. Sci., Vol. 2, No. 1) appears to have seen them in the blood of cattle infected with *Trypanosoma indicinum*, and to regard them as forming part of the developmental cycle. Holmes (Journal of Comparative Anatomy and Therapy, Vol. 17, 1904) figures in detail the formation of precisely similar bodies in the case of *Trypanosoma evansi*, and regards these bodies as true portions of the life cycle, the illustration of the details of the formation of the latent bodies in this paper being extremely accurate. The latent bodies we have dealt with in *Trypanosoma gambiense* probably also correspond to the bodies figured by Prowazek in the rat louse.

was still possible that they might eventually simply disappear altogether, the nuclear constituents of the trypanosome body being perhaps more resistant than the rest of the protoplasmic structure of the cell.

In order to throw some light upon this matter, we examined a number of infected animals which had been treated with atoxyl, this substance having the effect of destroying the parasites in the blood in the course of a very few hours at the most. When the blood of such animals was treated by injection of atoxyl and examined during the time when the parasites were still increasing in number, it was found that a large percentage of the trypanosomes could be observed dead among the corpuscles of the blood, but during the rapid disintegration which follows nothing comparable to the formation of the latent bodies is encountered. On the contrary, the nuclei in these instances are among the first of the organs to be affected by a general disintegration, which rapidly produces masses of débris, wherein it is only just possible to recognise from their shapes the remains of trypanosome cells (see fig. 41).

The same appearance is produced through the disintegration and death of the trypanosomes in the blood of an animal which has been killed by the disease.

From all this, it would appear that the latent bodies are not produced during the ordinary course of cell disintegration, and must be considered from some other point of view.

At this point it is, however, necessary to explain that when animals are injected with atoxyl at a time when the trypanosomes are decreasing in numbers in the blood, the disintegration does not necessarily overtake all the trypanosomes present. It is found, in fact, that a certain number of trypanosomes under such conditions do not succumb to the effects of the drug, but round themselves up and become encysted (see figs. 37, 40). These cysts are, however, very much larger than the latent bodies. They appear to be true resistant forms produced directly in response to the drug, and are not in any way comparable to the latent bodies we have just described.

With regard to the latent bodies which are produced at the maxima of an oscillating infection (structures which at first sight might, and probably would, be taken for disintegration products), these are, as we have seen, eventually collected in the spleen and

the bone marrow, and do not necessarily degenerate there. They persist in such situations in very large numbers, and each consists of a flattened nucleus with an intra-nuclear centrosome. There is also a vesicle attached to the nucleus, and the whole nuclear apparatus is surmounted by a thin film of protoplasm, figs. 27, 28.

At the periods when there are no trypanosomes to be found in the blood, these peculiar latent bodies are all the evidence of the existence of the parasites in an infected animal to be detected microscopically.

At the period of the infection when a few parasites begin to reappear in the blood, it is possible to still find numbers of latent bodies in the spleen, and in the bone marrow, wherein the intra-nuclear centrosome has divided into two, fig. 28.

Again, at this period it is possible to find forms in which one-half of the dividing intra-nuclear centrosome has passed out of the nucleus, fig. 28, forming an extra-nuclear centrosome. Still further, at a later period, we find forms in which a short flagellum has grown from the extra-nuclear centrosome, and these bodies subsequently appear to gradually transform themselves into small trypanosomes in the manner represented in figs. 28 to 32.

As the latent bodies are gradually transformed into small trypanosomes, the number of these bodies in the spleen and bone marrow diminishes, but it appears to be the case that a proportion of what are apparently latent bodies never really develop into trypanosomes and disappear altogether, or, in other words, that only a proportion of these bodies are under the circumstances above described capable of surviving the negative period, and once more forming themselves into complete trypanosomes.

The changes which we have now described are all those which we have hitherto been able to detect in relation to the succeeding stages of the infection in rats.

At first it was anticipated that further changes might have been encountered during different periods of the day and night, but although we have had the parasites in various animals under observation continuously at all periods of the curve of infection throughout several days and nights, no regular nocturnal alteration was discernible. However, it was found that the rapid diminution of the number of parasites almost invariably took place between 2 and 5 a.m.

So far as the above observations upon the life cycle of the

parasite of sleeping sickness have been carried, they appear thus to indicate a complete cycle in the blood of a single host, and the stages of such a life cycle can be semidiagrammatically represented in the manner given on page 445.

(C) THE MORPHOLOGY AND THE MULTIPLICATION IN THE BLOOD OF TRYPANOSOMA BRUCEI

The appearance of *Trypanosoma brucei* in the blood is represented in figs. 42, 43, 44, 45, 46. The chief morphological distinctions which the parasite of the disease Nagana presents when compared with *Trypanosoma gambiense* are found in relation to the distribution of the nuclear substance, and the characters of the extra-nuclear centrosomes.

The division of this trypanosome in the blood is longitudinal, as in the case of *Trypanosoma gambiense*. The nucleus divides amitotically. The division being first marked by a lengthening of the extra-nuclear centrosome until this body finally separates, and to two minute beads. At the same time the nuclear substance also elongates until we observe forms such as those represented in figs. 43, 44, 45, 46.

As in *Trypanosoma gambiense*, the stages of the division of the extra-nuclear centrosome and that of the nucleus may not be the same, at any particular time, and through this circumstance we observe the same sort of multiple, and abnormal forms as in the case of *Trypanosoma gambiense*.

(D) THE MORPHOLOGY AND THE MULTIPLICATION IN THE BLOOD OF TRYPANOSOMA EQUINUM

In the blood, *Trypanosoma equinum* possesses much the same shape as that of either *Trypanosoma gambiense* or *Trypanosoma brucei*; the nucleus is, however, usually placed nearer to the broad end of the cell. The extra-nuclear centrosome is large, and the nuclear division which takes place during the fission of the cells possesses points of much interest, since the centrosomes are more conspicuous in *Trypanosoma equinum* than in many other forms we have examined. The changes which take place in the intra-nuclear centrosome during the division of the nucleus can be studied with great clearness.

When the nucleus is at rest the intra-nuclear centrosome is surrounded by a light space, which is in turn enclosed by the stainable nuclear substance (fig. 47). During division the intra-nuclear centrosome divides, as in figs. 48 to 52. The nuclear substance becoming at the same time collected up in the region of the dividing intra-nuclear beads. As this process continues, the nuclear substance eventually forms itself into two cup-shaped masses situated around, and beyond the intra-nuclear centrosomes. Owing to this, when *Trypanosoma equinum* is dried the edges of these cups becoming flattened down produce at least two irregular bands on each side of the intra-nuclear centrosomes, which under these circumstances may suggest the existence of nuclear chromosomes. This appearance is, however, misleading. The nuclear division of *Trypanosoma equinum* being, as can be seen from figs. 47 to 53, really amitotic, as in the case of *Trypanosoma gambiense* and *Trypanosoma brucei*.

(E) CONSIDERATION OF THE FOREGOING OBSERVATIONS

For the sake of convenience, it is desirable to consider the latent forms as the starting point in the life cycle of *Trypanosoma gambiense*. These bodies, which in an ordinary fluctuating infection may remain unchanged for long periods in the organs (and, to a less extent, in the blood), consist at first of a nucleus containing an intra-cellular centrosome. The nucleus is related to a vesicle, and the whole nuclear apparatus is surrounded by a delicate film of protoplasm. At a later stage, the intra-nuclear centrosome divides into two, and one of these bodies passes out of the nucleus into the outer layer of protoplasm, which gradually increases in extent.

The above process results in the formation of an extra-nuclear centrosome. The extra-nuclear centrosome, and nucleus together with the intra-nuclear centrosome, henceforth form two entirely distinct sets of structures, which remain distinct through a very long series of divisions, as represented in the diagram on page 445, under the letter B.

After the first separation of the extra-nuclear centrosome from nuclear apparatus, both these sets of structures multiply independently throughout the succeeding series of generations until the

period at which the black line is formed. At this period the extra-nuclear centrosome develops a bridge, as it were, and connects itself for the time with the nucleus. It may be assumed that during this period some substance from the extra-nuclear centrosome passes into the nucleus. Anyhow, after this has taken place the remains of the extra-nuclear centrosome are very shortly cast away, together with the greater part of the protoplasm forming the rest of the cell, and the old flagellum.

Thus, if we consider the nuclear apparatus in the latent body as a whole, this would seem to be divided into two parts during the development of the trypanosome. After the formation of the cell is complete, these two structures, the nucleus and the intra-nuclear centrosome, remain in the same state, and multiply independently into similarly distinct bodies contained in the cells produced by all the longitudinal fissions. In other words, there arises from a nucleus A, two new structures, B and C, both of which differ from A. B and C multiply independently as the animals divide, but at a subsequent stage a portion of each B unites again with the C in all the cells, and the condition of the organism immediately reverts to A once more.

We have thus, after the formation of the latent bodies, an unequal division of the nuclear apparatus of the latent body, so as to form two different sets of structures, the nucleus with one centrosome, and the other centrosome by itself. Each of these then multiplies indefinitely in number. In individual cells these structures subsequently unite temporarily, and later the nucleus characteristic of the latent body is produced once again. In other words, dissimilar structures are formed from a nucleus by division, both derivatives multiply by division, and after a time unite in pairs, and the first type of structure is again produced. There is in this process, when contemplated from the present standpoint, an obvious similarity to the two forms of sexual elements in the higher animals and plants; to two sorts of gametic nuclei, or to a sexual dimorphism. A dimorphism in the trypanosomes which is in like manner followed by a reunion, or conjugation between the dissimilar elements, and succeeded by a reversion to the conditions obtaining before the dimorphism was produced.

The procedure in the case of the trypanosome nuclear apparatus differs, however, from that of apparently all other known organisms

where the phenomena of sex are discernible, in that the dimorphic products into which the nuclear apparatus of the latent forms separate remain contained within the same animal during its successive fissiparous generation. With the exception of this difference, however, the phenomena observed are certainly comparable to the production of sexual gametes and their conjugation. In the forms with which we have been hitherto familiar, the retention of the nuclear apparatus related to both sexes in one cell, may thus be nothing more or less than a morphological curiosity, and in no way necessarily suggests that the process in the trypanosomes we have been considering is fundamentally different from an ordinary sexual differentiation.*

Assuming the phenomena with which we have just been concerned to be of the nature of a sexual process, still another view could be held with regard to them. It may be suggested that the metamorphoses connected with the appearance of the black line is an attempt on the part of the trypanosomes to become sexually differentiated, but this attempt is not completed, the cells reverting to their primary condition, in which case the process could be regarded as an example of a special form of parthenogenesis. In relation to the above suggestion, it should be noted that, at the time of the formation of the black line, the whole of the extra-nuclear centrosome does not reunite with the nucleus, but that only one portion of the extra-nuclear centrosome does so. One moiety of the extra-nuclear centrosome is detached from the other, and one of these portions with the flagellum disappears together with the cytoplasm forming the trypanosome body.

We thus start with the separation of an extra-nuclear centrosome from an intra-nuclear centrosome through the division of an original intra-nuclear centrosome in the latent body. But at the end of the cycle, the nucleus enters again into connection with the extra-nuclear centrosome, yet it only does so with regard to a part of this body. On the other hand, it may be that the extra-nuclear centrosome

*It should be noted that there is no nuclear reduction in this process. The intra-nuclear centrosome divides, and one half of this body passes out of the nucleus. Both nucleus and extra-nuclear centrosome then divide in the production of the succeeding fissions. Afterwards the extra-nuclear centrosome or a part of it re-unites with the nucleus, and the rest of the cell body disappears. It is known that during fertilization a centrosome is often brought in with the male element: to this extent the process of fertilization is similar to the above.

divides normally, and that it is the half so produced which enters into connection with the nucleus during the formation of the black line. In this case, it may be that the nucleus receives once more a morphologically complete extra-nuclear centrosome produced in the ordinary way by division. Our observations are not conclusive with regard to these matters.

Considering the whole question from another point of view, it should be remembered that it is only among a percentage of the trypanosomes present during the periods of the maximum numbers of the parasites in the blood which can be found at any time to exhibit the formation of the black line, and we have to assume either that all the trypanosomes which ultimately form the latent bodies have passed through this metamorphosis, or that only some of them have done so.

We have no conclusive evidence upon this question. If, however, it should eventually be shown that only a fraction of the latent bodies are produced from trypanosomes which have passed through the black line metamorphosis, then it will become clear that there are two series of latent bodies, one class produced after the black line formation, and the other without this taking place. Should this be the case, the suggestion that the black line metamorphosis represents a peculiar form of the sexual act, or conjugation, which we have merely provisionally considered, will be found probably to be inaccurate. There will be an actual dimorphism among the latent bodies, and it will in this case be strongly suggested that the actual sexual act has yet to be discovered, and has been overlooked. We think it would be unprofitable to pursue this question further in the present Memoir. It is obviously a question that can only be properly considered after the phases of the life cycles of other trypanosomes are available.

In the introductory portion of this paper it was pointed out that there exists a complete discrepancy between "reduction" as apparently understood by Schaudinn,* and reduction as understood by biologists in general. Without throwing any reflection whatever upon the correctness of Schaudinn's observations in relation to the phenomena exhibited in the life cycle of *Trypanosoma noctuæ*, it is clear that, whether the process he describes exists or not, this process

* Loc. cit.

has nothing to do with chromosome reduction as ordinarily understood. His conception is a resurrection of Balfour and Minot's idea regarding the function of the polar bodies in the egg, and supposed corresponding structures in the male cells, as machinery whereby the physical representative of the opposite sex is got rid of more or less completely before fertilization.

In our observations upon *Trypanosoma gambiense* and other forms, in which the life cycle is possibly a complete cycle within the body of one host (see diagram, page 449), we have encountered nothing at all resembling the process stated to take place in the life cycle of *Trypanosoma noctuæ*. Prowazek,* dealing with the morphology and life cycle of *Trypanosoma lewisi*, maintains that the sexual act (in the form of ordinary conjugation) may take place either in the body of the rat-louse, or in the blood of an infected animal—the latter more rarely. According to this author, conjugation is preceded by a reduction process, which he describes in the following words:—

"Wie bereits erwähnt wurde, findet auf den mittleren Stadien
"der Verdauung in Mitteldarm die Reduktion der Flagellaten statt,
"die aber nicht gleichzeitig die beiden Kerne, den Blepharoplast und
"den centralen Kern, erfasst; bald ist der letztere schon völlig
"reduziert, während der erstere erst in den Prophasen dieser
"Vorgange steht und umgekehrt.

"Im centralen Kern wird vor der Reduktion zunächst das
"Karyosom bedeutend deutlicher und intensiver farbbar, das
"Chromatin wird körnig doch vereinigen sich bald diese Körner zu
"einzelnen Strängen (Taf. II, fig. 23, 24), die schliesslich nach Art
"von vier Reifen das inzwischen geteilte Karyosom umgeben. Dieses
"Stadium möchte ich mit den Stadien der Chromosomen Paarung
"der ersten Teilung der Metazoenspermatogenese vergleichen (Taf.
"fig. 24). Spater findet man in Kernhohlraum wiederum acht
"zerstreut liegende Chromosomen (Taf. II, fig. 26). Ein Stadium
"Vierergruppenbildung ist nicht sehr deutlich ausgebildet obgleich
"Andeutungen in diesem Sinne vorhanden waren (Taf. II, fig. 25).
"doch kann man wegen der Kleinheit des Objektes nichts sicheres
"diesbezüglich aussagen, obzwar in dem abgebildeten Fall doch

* Prowazek, loc. cit., page 372.

" 16 Chromosomen, die durch die zwei Teilungen auf vier reduziert " werden, gezählt werden könnten. Deutlicher waren die Bilder bei " *Herpetomonas*. Durch die endlich effektiv gewordene Teilung des " Karyosoms wird der erste Reduktionskorper gebildet, der selten als " ein dunkles, körniges Gebilde gegen das spitze Ende der *Z*elle " abdrückt, sondern meistens dicht am Kern selbst liegen bleibt " (Taf. II, fig. 30). Bald darauf vollzieht sich noch eine Teilung, " durch die der zweite Reduktionskorper gebildet wird. In fig. 31 der " Taf. II, bemerkt man terminalwärts diesen Reduktionskern, der ein " kleines Karyosom und die vier dicht verbackenen Chromosomen " enthält. Demnach muss der reduzierte Kern nur vier Chromosomen " besitzen. An dem sich reduzierenden Blepharoplast kann man " nicht so viel Details erkennen ; zunächst teilt sich der Blepharoplast " in zwei Teile (Taf. II, fig. 29), von denen der eine Teil durch eine " heteropole Spindel noch einer Reduktionsteilung unterliegt. Der " este Reduktionskorper übernimmt manchmal die undulierende " Saumgeissel und degeneriert erst ziemlich spät. In anderen Fallen " bleibt die Geissel an dem reduzierten Blepharoplast haften (Taf. II, " fig. 28, 29)."

Prowazek describes the nuclear division taking place during the fission of the parasites in the blood in infections with *T. brucei* and *T. lewisi* as being mitotic, and the nuclei as containing eight chromosomes. Our observations upon *T. brucei* and the other trypanosomes with which we have been working, are all similar in regard to this matter. When these animals have been properly preserved there appear to be no chromosomes produced, and the type of nuclear division during the fission of the animals is invariably amitotic, the extra nuclear centrosome and the nuclear substance dividing like drops. It is possible, of course, as in the case of the Ciliata, that the divisions become characteristically mitotic immediately before reproduction, but in none of the trypanosomes which we have examined (one of which, *T. equiperdum*, certainly runs its life cycle in a single host) have we encountered anything of the kind.

We have had these forms under continuous observation for more than a year, and for long periods at all hours of the day and night In the case of *Trypanosoma equinum*, we were at first inclined tc think that the division was of the mitotic type, but this inference was soon found to be simply due to the defective manner in which the

animals had been preserved and dried, or to other forms of indifferent fixation. In other words, there are often produced during fission of the animals under such circumstances appearances in their nuclei due to coagulation effects which may readily be mistaken for nuclear chromosomes. As the fixation becomes better, in all the forms with which we have hitherto dealt, such appearances can, however, be clearly shown to be illusive, and the division of the nucleus during fission to be invariably amitotic in character. The same inference is borne out by a study of the living animals.

Among the illustrations given by Minchin [Proceedings of the Royal Society, Vol. 78, Series B, No. 20, Plate 12], figs. 4, 8, 9, and 17 have been produced from specimens that have been dried and stained, and suggest the existence of chromosomes, but we are inclined to think that these appearances are simply due to the bad fixation methods employed, and are really quite misleading.

With regard to the nature of the nuclear division accompanying fission of the above-mentioned trypanosomes, our results are in complete accord with those of Laveran. Indeed, with regard to the nuclear reduction described by Schaudinn,* and finally by Prowazek, our observations have revealed nothing suggesting anything even analogous to these descriptions. Prowazek gives a series of figures illustrating this process in *Trypanosoma lewisi* (Pl. II, figs. 23, 24, 25). Here nuclei with eight chromosomes are said to be apparent. We cannot detect in the figures themselves the slightest suggestion of this being the case, and are inclined to think that the irregular blotches and strands, undoubtedly correctly drawn, have nothing to do with chromosomes, but are due to the manner in which the specimens have been preserved. In figs. 27, 28, 29, 30 and 31, the so-called reduction of the nucleus as well as the extra-nuclear centrosome (blepharoplast) is represented; but in none of these figures do we see any indication of either true chromosomes, true mitotic division, or true reduction as ordinarily understood. In fact, as far as the illustrations are concerned, we are unable to find, or to see, any indication of a reduction process.

It will thus be observed that the results we have obtained, especially in relation to *Trypanosoma gambiense*, but also equally

* Generations und Wirtswechsel, &c.

† Loc. cit.

among other forms to be described later, differ not only in degree but also in kind from those obtained on the one hand by Schaudinn, and on the other by Prowazek. The descriptions of Schaudinn, in so far as they bear at all upon the present work, do so, however, through the investigations of *Trypanosoma noctuæ*, which since it appears to be a form of trypanosome requiring more than one host for the completion of its life cycle, may very likely differ in the features of this life cycle from the more ordinary forms with which we have been concerned. On the other hand, Prowazek's results obtained in the case of *Nagana* (that is to say, from one of the trypanosomes considered in the present paper), so far as the nuclear changes during fission are concerned, differ entirely from our own; these latter fall directly into line with the observations we have made upon other forms, and are quite incompatible with the description of this process given by Prowazek in the case of *T. lewisi* or *T. brucei*. The question which now confronts us is upon what cause this difference of results depends. We are inclined to think that the difference of result is due to the methods which have been employed. We may as well say here, that from what we have gathered with respect to the different methods that have been generally in use, it appears that all the methods involving the drying of the blood before staining, or, in fact, any method involving drying at all, is, so far as nuclei are concerned, absolutely useless from a cytological point of view. Nothing relating to the delicate mechanism of mitotic division is generally preserved in cells, whether they belong to unicellular or multicellular organisms, when dried and stained with Romanowsky, Giemsa, or in any other manner. Even the resting nucleus itself under such conditions becomes a mere caricature of the actual structure.

When treated in this way, the irregular or regular blotches and streaks of stainable matter have nothing in common with, and do not represent, even in a relative or equivalent sense, the structures actually present in the cells. Anyone who wishes to verify this fact for himself will have no trouble in doing so if, for example, he makes a smear preparation from the testis of a rat, stains after the manner of Romanowsky, and then compares this with a properly fixed and stained smear, in the production of which ordinary cytological precautions have been observed. It is a curious fact that in a rat's testes under these conditions certain cells which really contain

sixteen *gemini* or heterotype chromosomes, when subject to the action of drying and Romanowsky, very often present (within the ill-formed area representing the nucleus) six irregular masses of stainable stuff resembling the so-called chromosomes of the dried trypanosomes. Such appearances are, however, certainly due to regularity of coagulation and shrinking during the drying of the cells, and have nothing in common with the real morphological structures, (the chromosomes), which either the living cells, or successfully-preserved cells possess. In view of these circumstances, we are inclined to regard many accounts of the existence of chromosomes, spindles, and even the assumed existence of mitotic division among trypanosomes, as conclusions which appear to be most questionable, and as requiring in all cases confirmation in a variety of ways which do not involve the violence dealt to the finer details of cell structure by drying.

Finally, we see in the case of *Trypanosoma gambiense* that the life history of this parasite as it lives in rats seems to be complete in the blood of the rat, and not in any way dependent for its completion upon the transference of the parasites into the blood of any other kind of host. In rats the latent forms pass gradually into trypanosomes, these in turn divide through many generations, and their multiplication is followed by a metamorphosis which, whether we regard it as a special form of sexual process, as a form of pathogenesis, or as a sexual stage, the fuller details of which have not yet been elucidated, seems undoubtedly to stand in one of these relationships to the normal cell multiplications preceding the formation of latent bodies. The stage in question results in the production of the latent bodies once more, and the cycle is complete.

It may be objected to this conception that, notwithstanding the cyclic development of *Trypanosoma gambiense*, still there may exist a possibility, or probability, of the transference of the trypanosomes into some other host where a further metamorphosis representing the sexual stage of the organisms is passed through. This, of course, may be so, but we have in the case of the trypanosome of *Dourine* a clear instance of a trypanosome life history, which, under normal circumstances, is not transferred into any other kind of host; and, under normal circumstances, *Trypanosoma equiperdum* must pass through whatever sexual stage it may possess, its whole life history in fact, in

the body of the horse. *Dourine* can, however, like sleeping sickness, be inoculated from host to host by simple transmission of blood as well as by coitus; in other words, the faculty of being transmitted by simple inoculation of blood is shared by *Trypanosoma equiperdum*, wherein no other host is usually involved, as well as by *Trypanosoma gambiense*. In these circumstances, it is simply natural, assuming flies to be the agents by which sleeping sickness is transmitted, to admit that this form of transmission may be merely in the nature of a mechanical transference, and have no more relation to the sexual act in the life cycle than has the artificial withdrawal of blood from a horse infected with *Dourine*. In other words, it would seem that the transference by flies in the case of sleeping sickness may have no more significance with respect to the life history of the parasite than has the direct inoculation of *Dourine* from horse to horse by means of a needle.

As we have already pointed out, the observations of Bruce, Dutton, Todd and Hanington* and others seem to indicate that the transference of sleeping sickness, when it is brought about by flies, is in the nature of a simple inoculation of blood, while it would appear that Dutton, Todd and Hanington incline further to believe that flies are not necessarily the normal means by which the propagation of sleeping sickness takes place.

They sum up the situation in this respect as follows:—"It seems "that all the results are in conformity with the hypothesis that "*Glossina palpalis* transmits *Trypanosoma gambiense*, and that it "is probably not able to do so when the space between the trans- "mitting feeds exceeds 48 hours; this conclusion is, nevertheless, to "our minds a most unsatisfactory one, if we are to regard these "Glossinae as the chief or only carriers of *Trypanosoma gambiense*. " . . . It certainly seems possible that mechanical transmission "by tsetse flies cannot alone be responsible for the rapid spread of "sleeping sickness of recent years."

These questions, however, open out a wide field of enquiry, which it is at present unprofitable further to discuss.

* Loc. cit.

APPENDIX I

METHOD OF PREPARING AND STAINING WET FILMS, USED DURING THE FOREGOING INVESTIGATIONS

Place a very thin layer of albumen-glycerine on a clean slide. The best method is perhaps to put a drop the size of a large pin's head on the slide, and to spread this with a clean duster over the slide. On top of this layer spread a drop of blood in the usual way, and dip the slide, while wet, into the fixing solution (Flemming's strong solution was usually used). Leave it for about five to ten minutes, wash immediately in water, and pass the slide through alcohols in consecutive order, increasing by 10 per cent. at a time to absolute alcohol. Then back from absolute into 80 per cent. alcohol in which is contained iodine and potassium iodide. In order to prepare the solution of 80 per cent. alcohol containing iodine and potassium iodide, make up a concentrated solution of potassium iodide in water, and iodine in alcohol, mix them together, and add them to some 80 per cent. alcohol until the mixture becomes a dark brown colour. Leave the slide in this from five to ten minutes, and then bring it into 30 per cent. alcohol. Use for staining either aniline safranine (nach Babes), or the following solution:—Prepare a concentrated watery solution of safranine (Grübler) and a concentrated alcoholic solution of safranin, mix them in equal parts, and then add pure aniline oil. Shake from time to time, and leave the solution to ripen for three to six months. Stain in this solution for from half an hour to two hours. Wash off the safranin, and stain afterwards with polychrome methylene blue [one gramme methylene blue purissimum medicinale (Höchst) 100 c.cm. distilled water and ·5 gramme sodium carbonate, left in an incubator to ripen; the older the solution the better the staining properties]. Wash off the methylene blue, and differentiate with Unna's orange tannin, as long as the blue stain comes out. Bring the slide up through alcohols, as above, into absolute alcohol. The film has then a reddish colour. Now dip the slide into aniline oil until the reddish colour changes to a purple-blue tinge; the aniline oil takes out at the same time the excess of blue stain left by the orange tannin. Clear in xylol. Mount in Canada Balsam under a coverslip.

APPENDIX II

MODIFICATION OF HEIDENHAIN'S HAEMATOXYLIN METHOD, USED DURING THE FOREGOING INVESTIGATIONS

Fix and treat the film in the way described in Appendix I. Clear the slide from the alcohol containing iodine and potassium iodide, and pass it through successive alcohols (as in Appendix I) into water. Stain in a $3\frac{1}{2}$ per cent. solution of iron alum for one hour, wash this off, and stain with the following solution:—·5 gramme haematoxylin dissolved in 100 c.cm. distilled water, to which, after the haematoxylin has dissolved, a few drops of concentrated watery solution of lithium carbonate is added. Stain for half an hour, and then differentiate in the usual way with iron alum.

DESCRIPTION OF FIGURES

In all cases, unless otherwise stated, the figures have been drawn with a long tube Zeiss, 2 mm. apo. objective and 18 or 27 eyepiece.

PLATE XXXVIII

Trypanosoma gambiense

Figs. 1, 2, 3 from peripheral circulation stained with iron haematoxylin.

Figs. 4 and 5 stained with Breinl's stain.

Fig. 1.—Trypanosome in the resting condition. Nucleus single, not dividing. Intra-nuclear centrosome single.

Fig. 2.—Trypanosome showing early stages in division of nucleus, intra-nuclear centrosome divided; extra-nuclear centrosome also divided, new flagellum growing.

Fig. 3.—Trypanosome in the same stage as preceding. Intra-nuclear centrosome completely divided; the new flagellum is also seen during the course of its development.

Fig. 4.—Trypanosome is in the same condition as fig. 1, showing also the metabolic granules in different parts of the cell.

Fig. 5.—Trypanosome wherein two undulating membranes have been formed, showing also the metabolic granules.

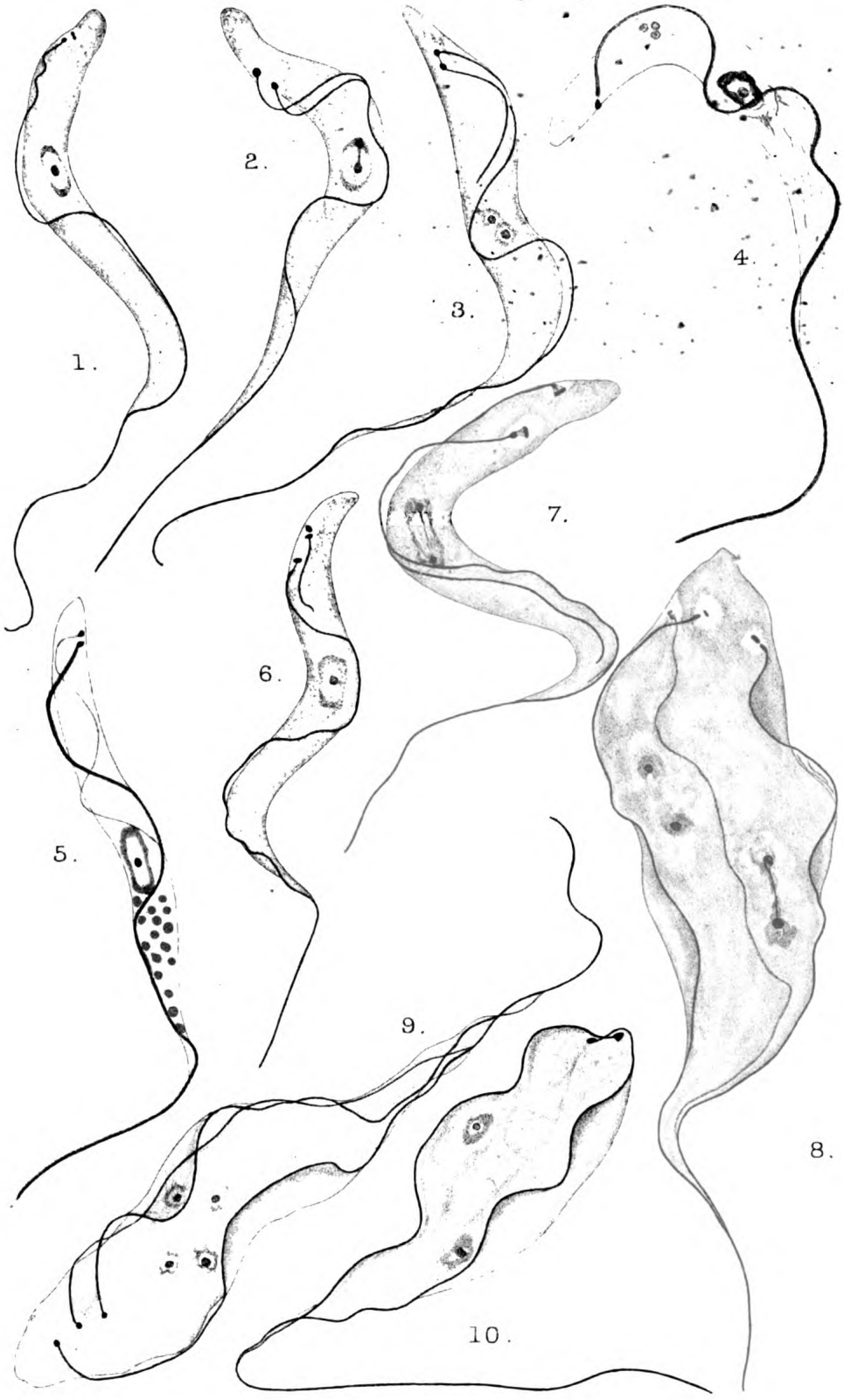
Fig. 6.—Trypanosome in much the same condition as in fig. 1, but showing early stages in development of the new flagellum.

Fig. 7.—Later stage in the division, showing the mode of division of the intra-nuclear centrosome, amitotic fission of the rest of the nucleus, and the duplication of the extra-nuclear centrosome.

Fig. 8.—Trypanosome showing three flagella and three nuclei in different stages of division.

Fig. 9.—The same showing three flagella and four nuclei.

Fig. 10.—The same showing two flagella and two nuclei.



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PLATE XXXIX

Trypanosoma gambiense

Figs. 11 to 14 stained with Breinl's stain.
Figs. 15 to 21 stained with the modification of Heidenhain's haematoxylin.

Fig. 11.—Trypanosome showing division of the intra-nuclear centrosome, and the nuclear substance. Also multiplication of the intra-nuclear centrosome so as to form a group.

Fig. 12.—Trypanosome showing mode of amitotic separation of the nuclei. Multiplication of the intra-nuclear centrosome, and the formation of an independent group of these bodies. In this and the preceding figures metabolic granules are also seen.

Fig. 13.—Later stage in the fission of a trypanosome. The flagella are being detached from one another at the thin end of the cell. The cell body is dividing from this end towards the other. The nuclei are already divided. Metabolic granules are scattered throughout the spongioplasm.

Fig. 14.—Later stage in the division of a trypanosome, showing the manner in which the daughter cells separate.

Fig. 15.—Trypanosome, at one of the maximum periods of the infection, showing a single flagellum and resting nucleus, and also the origin of the black line from the extra-nuclear centrosome.

Fig. 16.—A similar stage wherein the black line has reached the neighbourhood of the nucleus.

Fig. 17.—The same.

Fig. 18.—Trypanosome showing the black line coiled upon itself towards the intra-nuclear centrosome.

Fig. 19.—Trypanosome showing early stages in the degeneration of the black line, and its later direct association with the nucleus.

Fig. 20.—Similar stage, wherein the intra-nuclear centrosome has become divided.

Fig. 21.—So-called involution stage, showing resting nucleus and multiplication of the intra-nuclear centrosome.

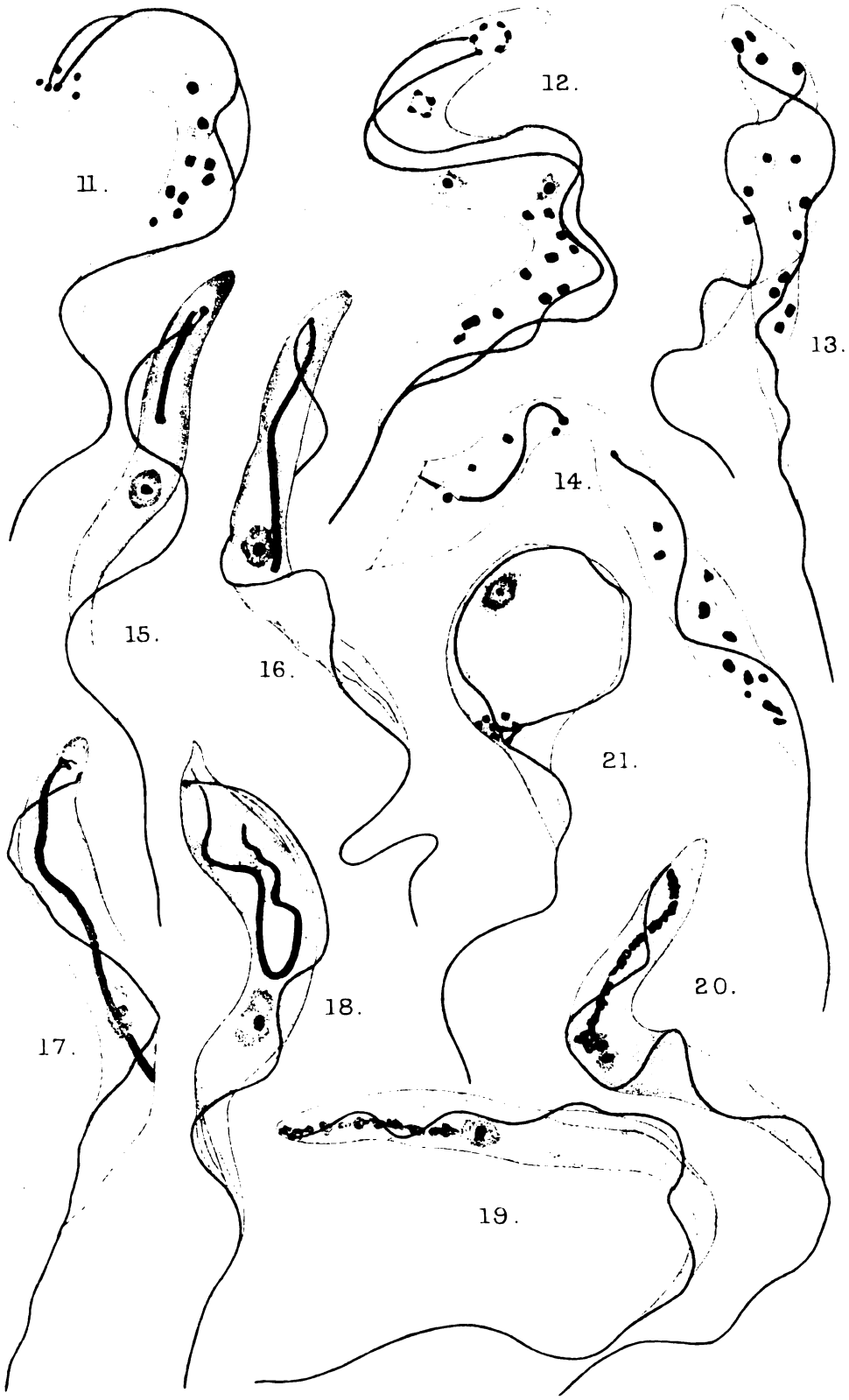

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PLATE XL

Stages in the Metamorphosis of Trypanosoma gambiense in the Organs

Figs. 22, 23, 24, 26, 27, 28, 29, 30, 31, 32, 34, 35, 36, stained with a modification of Heidenhain.

Figs. 25 and 33 stained with Breinl.

Fig. 22.—Trypanosome during the decrease of the parasites in the blood of a rat, showing alteration in the nucleus marked by the formation of a vacuole.

Fig. 23.—The same.

Fig. 24.—One of the common forms in the lung at this period, showing the same changes in the nucleus.

Fig. 26.—Low power view of trypanosome at this period, showing the detachment of the latent body from the parent cell.

Fig. 27.—Latent bodies. The nucleus is attached to a vacuole and both are surrounded by a thin film of protoplasm. The nucleus contains a single intra-nuclear centrosome.

Fig. 28.—Latent bodies. To the left the intra-nuclear centrosome is shown divided. To the right stages in the division of this body and the extrusion of one daughter element from the nucleus.

Fig. 29.—Latent bodies showing origin of a new flagellum from the intra-nuclear centrosome.

Figs. 30, 31.—Later stages in the formation of small trypanosomes from the latent bodies.

Fig. 32.—Latent body from the spleen of a rat infected with *Trypanosoma brucei*, showing nucleus, vacuole, and the origin of the flagellum from the intra-nuclear centrosome. (Compare fig. 29.)

Fig. 33.—Trypanosome showing early stages in division of the nucleus.

Fig. 34.—Trypanosome drawn to show the Schaumplasma structure of the protoplasm.

Figs. 34 to 36.—The same.

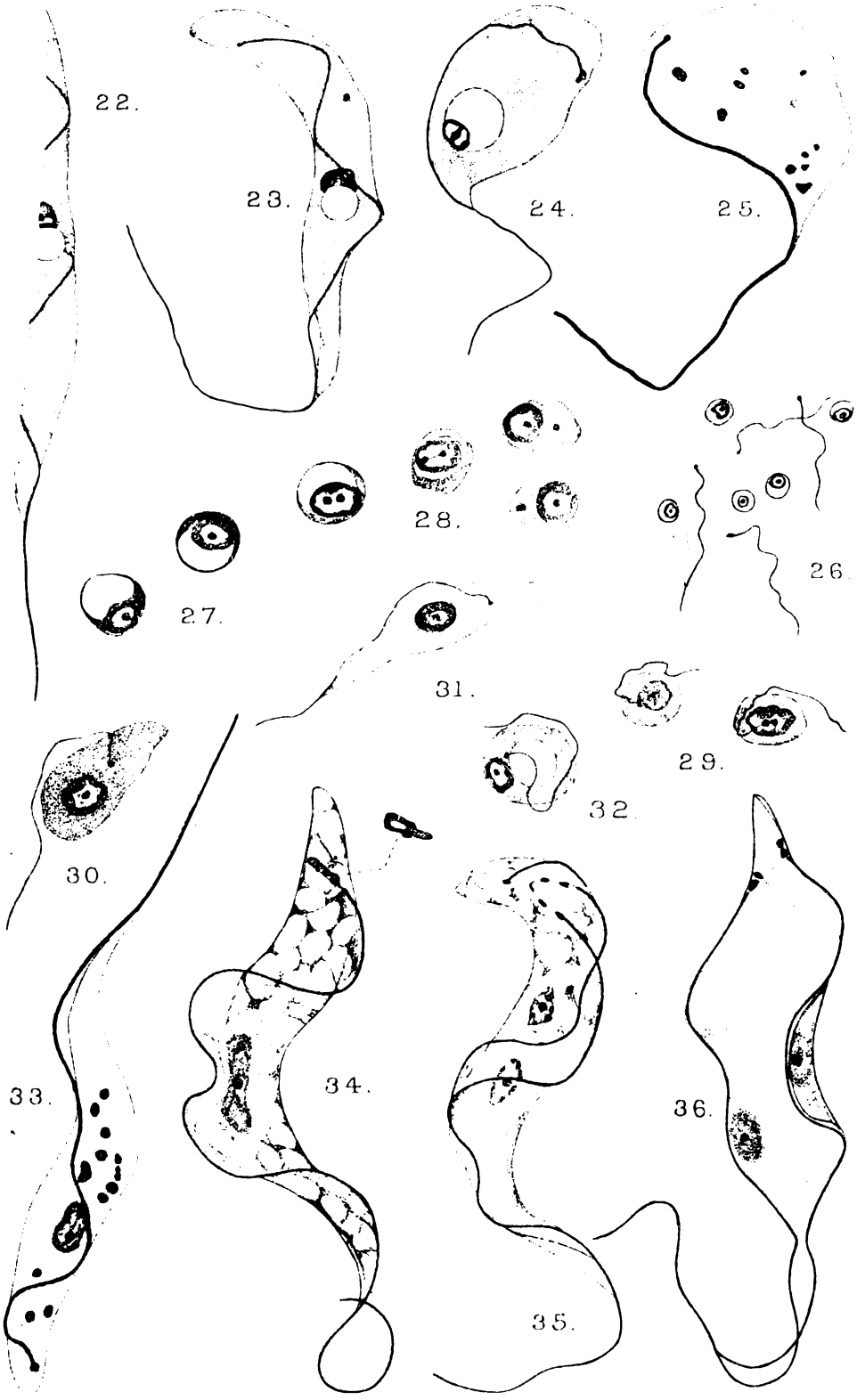

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PLATE XLI

Trypanosoma gambiense and Trypanosoma brucei

Figs. 37-41, *T. gambiense.*
Figs. 42-46, *T. brucei.*
Figs. 37, 38, 40, 41, 42, 43, 45, 46, Breinl's stain.
Fig. 44, Heidenhain.

Fig. 37.—Trypanosome in the blood of a rat after treatment with atoxyl, showing rounding up of the cell body.

Fig. 38.—Further stage in this process, flagellum is still attached. A slight modification of a membrane is apparent round the periphery of the cell.

Fig. 39.—The same, flagellum not visible.

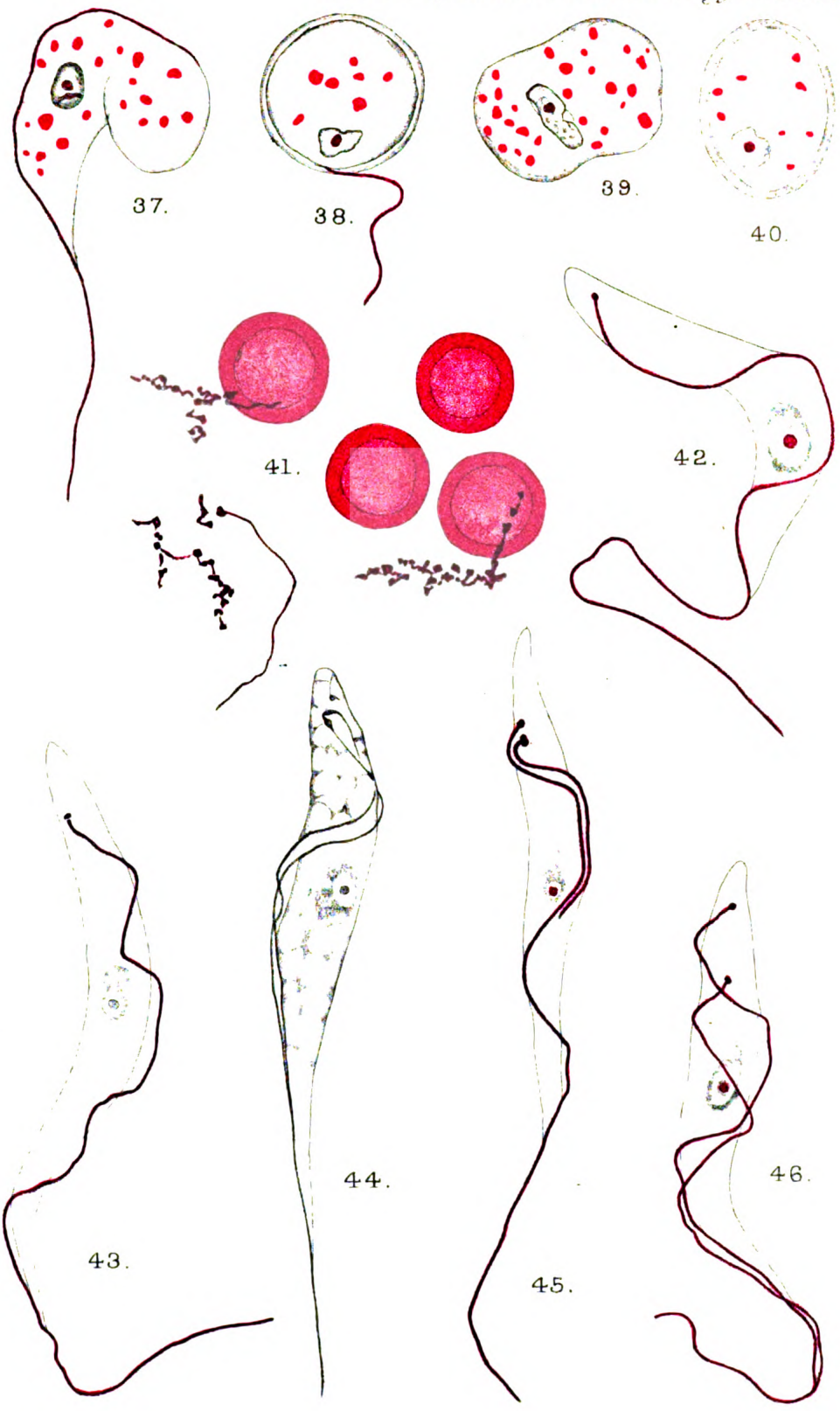
Fig. 40.—Later stage in the formation of the cyst. The membrane more apparent.

Fig. 41.—Trypanosomes killed by atoxyl in the blood.

Fig. 42.—*Trypanosoma brucei* in resting condition, showing structure of the nucleus and relation of the schaumplasm.

Fig. 43.—The same, showing the division of the extra-nuclear centrosome.

Figs. 44 to 46.—Later stages of division.



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PLATE XLII

Trybanosoma equinum

Figs. 47, 49, 50, 51, 52, 53, 54, stained with Breinl.
Fig. 48 stained with modification of Heidenhain.

Fig. 47. Resting stage of the trypanosome.

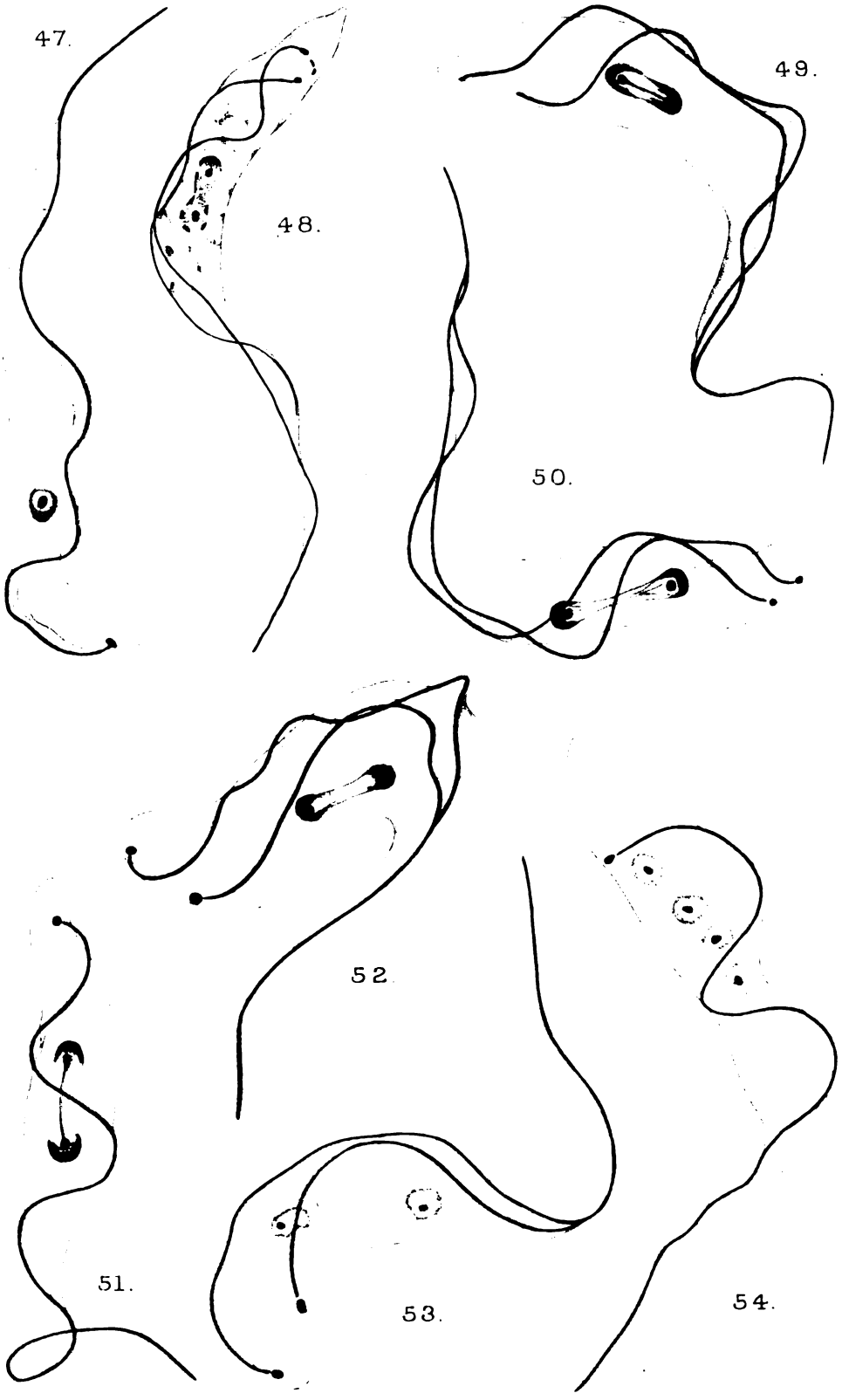
Fig. 48. Stage showing formation of the new flagellum and division of the nucleus.

Fig. 49. -Trypanosome showing details of the division of the intra-nuclear centrosome and the nuclear substance.

Figs. 50, 51. Trypanosomes showing later stages of the same process.

Figs. 52, 53. Trypanosomes showing still later stages in the division of the nucleus and the characters of the intra-nuclear centrosomes.

Fig. 54. -Trypanosome wherein the nucleus has divided into four constituents, although there is only one flagellum.



J. E. S. M. ad nat. del. Huth, Lith^r London

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ANNALS

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Regarding the Courses of Instruction proposed to be given by the Liverpool School of Tropical Medicine, and the Examinations for the Diploma of Tropical Medicine arranged to be held by the University of Liverpool during 1908 (subject to such alteration as may hereinafter be decided upon),

Lent Term begins January 14.
Lent Examination, March 23.
Summer Term begins May 1.
Summer Examination, July 13.
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The full Course of Instruction is open to all qualified medical men, and the examination to all students who have taken out this full course.

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1904	Augustine, Henry Joshua
1906	Bate, John Brabant
1904	Bennett, Arthur King
1906	Bennetts, Harold Graves
1907	Bond, Ashton
1907	Branch, Stanley
1905	Brown, Alexander
1904	Bruce, William James
1904	Byrne, John Scott
1905	Caldwell, Thomas Cathcart
1906	Carter, Robert Markham
1906	Chisholm, James Alexander
1904	Clayton, Thomas Morrison
1906	Clements, Robert William
1907	Collinson, Walter Julius
1905	Critien, Attilio
1904	Dalziel, John McEwen
1907	Davey, John Bernard
1904	Dee, Peter
1907	Donaldson, Anson Scott
1906	Dundas, James
1906	Faichnie, Norman
1907	Fell, Matthew Henry Gregson
1907	Gann, Thomas William Francis
1907	Graham, James Drummond
1904	Greenidge, Oliver Campbell
1904	Hehir, Patrick
1907	Hiscock, Robert Carroll
1905	Hooton, Alfred
1905	Hudson, Charles Tilson
1905	Illington, Edmund Moritz
1906	Jeffreys, Herbert Castelman
1907	Keane, Joseph Gerald
1907	Kennan, Richard Henry
1907	Kenrick, William Hamilton
1904	Khan, Saiduzzafor
1904	Laurie, Robert
1907	Le Fanu, George Ernest Hugh
1905	Macfarlane, Robert Maxwell
1906	Mackenzie, Donald Francis
1907	Mackey, Charles
1904	Maclurkin, Alfred Robert
1905	Maddock, Edward Cecil Gordon
1907	Maddox, Ralph Henry
1907	McCarthy, John McDonald
1904	McConnell, Robert Ernest
1905	Moore, James Jackson
1904	Nicholson, James Edward
1905	Nightingale, Samuel Shore
1906	Pailthorpe, Mary Elizabeth
1906	Palmer, Harold Thornbury
1906	Pearse, Albert
1904	Philipson, Nicholas
1905	Radcliffe, Percy Alexander Hurst
1907	Raikes, Cuthbert Taunton
1907	Ryan, Joseph Charles
1906	Sampey, Alexander William
1904	Sharman, Eric Harding
1906	Smithson, Arthur Ernest
1906	Taylor, Joseph van Someron
1906	Taylor, William Irwin
1904	Thomson, Frank Wyville
1906	Tynan, Edward Joseph
1907	Vallance, Hugh
1904	Walker, George Francis Clegg
1906	Watson, Cecil Francis
1906	Willcocks, Roger Durant
1906	Williamson, George Alexander
1905	Young, John Cameron

EDITORIAL NOTICE

By order of the Committee of the Incorporated Liverpool School of Tropical Medicine, the series of the Reports of the School, which have been issued since 1899, will be followed, from January 1, 1907, by the Annals of Tropical Medicine and Parasitology, of which this is the fourth number.

The Annals are issued by the Committee of the School, and will contain all such matter as was formerly printed in the Reports—that is to say, accounts of the various expeditions of the School and of the scientific work done in its laboratories at the University of Liverpool and at Runcorn. Altogether twenty-one Memoirs, besides other works, have been published by the School since 1899, and of these ten, containing 519 quarto or octavo pages and 95 plates and figures, were published during the two years 1904 and 1905; and there is no reason to suppose that this rate of production by the workers of the School alone will diminish in the future. In addition, however, to School work, original articles from outside on any subject connected with Tropical Medicine or Hygiene may be published if found suitable (see notice on back of cover); so that, in all probability, not less than four numbers of the Annals will be issued annually. Each number will be brought out when material sufficient for it has been accumulated.



OBSERVATIONS ON THE SO-CALLED 'CANARY FEVER'

OBSERVATIONS ON THE SO-CALLED 'CANARY FEVER'

BY

C. E. WALKER

ASSISTANT DIRECTOR OF CANCER RESEARCH AND HONORARY LECTURER ON CYTOLOGY TO THE LIVERPOOL SCHOOL OF TROPICAL MEDICINE IN THE UNIVERSITY OF LIVERPOOL

(Received for publication October 4th, 1907)

Hitherto the evidence regarding the disease commonly known as 'Canary Fever,' has been of a most conflicting character. Very frequently the disorder is of a comparatively trifling nature, and such cases do not usually come to the knowledge of a medical man. Private enquiry points to the probability that from 60 to 80 per cent. of the visitors to the Island suffer from one or more attacks during the time that they are in the islands. Dr. Taylor, who has practised among the English visitors in Las Palmas for many years, estimates that only about 5 per cent. of the cases that occur come under the observation of a medical man.

The disease is apparently more prevalent during the winter months than during the summer, but this is probably due to the fact that larger quantities of food are consumed during the former period owing to the larger number of visitors, and that as flies are then more numerous, there is, as we shall see later, a much greater chance of the food becoming infected.

Clinically the disease is characterised by the suddenness of its onset, and by its generally attacking a number of individuals in the same hotel at the same time. Frequently it commences with vomiting, and as a rule, even when vomiting does not actually occur, there is a considerable feeling of nausea which lasts for from a few hours to two or three days. A few hours after the vomiting or nausea, sometimes synchronously with it, griping pains in the abdomen of a violent character commence. This is the most marked and constant symptom, and causes very considerable suffering. The patient now developes a more or less acute attack of diarrhoea. In the more acute

cases the discharge frequently becomes mucous in character, and is sometimes blood-stained. Sometimes the temperature rises, but not, as a rule, to any considerable extent. The length of the attack varies in different cases; a slight one but for two or three days, the more serious for two or three weeks.

The disorder has been attributed to various causes. The red wine of the country, the water, an excess of fruit, too much food, and too little exercise have all been blamed. The most superficial examination of the available evidence, however, shows that none of these can be the usual cause of the disease. Individuals who are teetotalers are as frequently attacked as those who never drink water. Those who eat a quantity of fruit are no more subject to it than those who abstain from fruit altogether.

Enquiries made with regard to the milk supply showed that the dairies in the town of Las Palmas, where the cows are stall-fed, are generally remarkably clean. These dairies do not, however, supply enough milk to meet the demands of visitors, and a large proportion of it is brought down from the country. The milk supply is generally the same in the case of the hotels as it is in the case of the private houses, which fact, as we shall see later, is of special significance. Also I was able to ascertain that people who drank large quantities of milk in private houses were not subject to the disease, while those in the hotels who never touched milk were subject to it.

With regard to the water supply, there is no evidence incriminating it, for people who have stayed in hotels with a certain water supply have suffered from the disease, while people in private houses with a water supply derived from the same source have been free from it.

The most significant fact which was ascertained at the commencement of the enquiry was, that while visitors at hotels so frequently contracted the disease, it was practically unknown among people living in private houses. Dr. Taylor informs me that among English people who live in private houses the disorder is very rare, and that in his experience it is confined to visitors at the hotels. This coincides entirely with the personal evidence I was able to obtain.

Dr. Millares also informs me that the disease, in his experience, is practically confined to the hotels.

Typhoid appears to be comparatively common among the lower classes of natives, but I was able to ascertain the occurrence of only

two cases during the past three years in hotels, and in both these there was very good evidence that the infection had occurred outside the hotels in which the men were staying.

These facts suggested the advisability of a comparison between the sources of food supply and the methods of keeping and cooking the food in hotels with those in private houses.

The meat for hotels, in some cases at any rate, is frequently frozen meat brought to the islands by ship. It often happens that this meat is more or less exposed on the quay for several hours—sometimes for more than twelve when a steamer arrives early in the night.

The larders in many hotels are not fly proof, and the food-stuff is not adequately protected from flies before and after cooking. In some cases it is not protected at all.

Soup in the hotels is frequently made in a stock-pot which is only emptied once or twice during the week.

In private houses the meat supply is generally from animals killed in the islands. Considerable precautions are usually taken to protect all food from flies. This precaution extends not only to keeping meat, fish, &c., in fly proof safes, but even dishes of fruit are often covered with muslin while on the table. Soups are made fresh every day, and a stock-pot is not used.

In some of the butchers' shops (in Las Palmas, for instance) one frequently sees meat exposed for sale which is not protected from flies. I have been unable to trace the destiny of this meat. In the case of the private houses where I was able to trace the source of the meat supply, I found that a servant was sent early in the morning to the market. This meat is stated to be freshly killed. It must also be observed that the evidence with regard to the occurrence of the disease which is here given applies only to the better class of natives, and not to the lower classes.

Flies are extremely numerous, particularly during the time of the year when most visitors are staying in the hotels. They always swarm to an extent which can but rarely happen, even in isolated cases, in more temperate climates. Again, the habits of the lower classes among the natives are such as give perhaps the greatest possible number of chances of infection of the food with various micro-organisms by means of the flies. For instance, the inhabitants usually defaecate in any spot that suits their personal convenience, and one spot

favoured particularly by them in this way is not many yards away from the unprotected larder window of one of the hotels. This instance is only quoted as one among a large number.

Temperature seems also to have a very marked effect upon the frequency of the disorder. In Las Palmas, for instance, the summer and winter temperatures vary only by about 10 degrees, and the temperature does not usually go below about 75°F., so that any bacteria carried on to food-stuff by flies would be under very favourable conditions for rapid multiplication. In the Monte district of the same island the temperature is much more variable, and during the winter is comparatively low. In this district cases of so-called Canary fever are comparatively rare in the hotels.

These observations indicate that there should be a greater risk of conveying a large dose of bacteria, and of the toxins they produce, by means of soup from the stock-pot and of rechauffés, than by any other kind of food. The soup is rarely if ever brought to boiling point, and necessarily is allowed to cool every night when the kitchen fire goes out. Entrées and other forms of rechauffés are made from meat or fish that has been previously cooked, and such material will have been left for from 18 to 24 hours before being served without having been brought up to the boiling point of water. Micro-organisms will have been destroyed, and many of their toxic products may have been disintegrated by the temperature generally required to cook food for the first time, and with such material the risk of infection or poisoning will be much less than in the case of the soup from the stock-pot or of a rechauffé which is rarely if ever brought up to a sufficiently high temperature.

During my stay in Las Palmas, most of the other visitors in the hotel had attacks of the disorder. My two companions and I avoided soups and entrées, and also avoided infection. On one occasion, however, one of my companions took an entrée. An attack followed within a few hours. This certainly might be interpreted as a coincidence, but when taken in conjunction with the fact that the disease is confined to the hotels, and keeping in mind the difference in the method of keeping food in the private houses, it might also be regarded as corroborative evidence.

The symptoms of the disorder, its epidemic character, and its being confined to hotels, suggest very strongly that it is due to

bacterial infection of the food. The details which have been given regarding the differences between the methods pursued in hotels and in private houses, suggests that the infection is largely brought about by flies carrying the bacteria on to the food, in which position the conditions are particularly favourable as regards temperature and nourishment for their rapid multiplication. This does not necessarily suggest any specific micro-organism, but is amply sufficient to account for all the cases of Canary fever that occur. There is, however, another cause which might explain some cases, which I have proved by personal experience to be sufficient to produce all the symptoms of the so-called fever. If two or three men live for many days upon a small boat during the summer months in England, they will very likely be suddenly attacked by these symptoms in an acute form unless all their cooking utensils, plates, forks, &c., be thoroughly cleaned and boiled at frequent intervals. The temperature below deck and in the lockers in a small boat in British waters will frequently be very high during the summer, and there will be very favourable conditions for the multiplication of bacteria in any small collections of grease, &c., on the cooking and other utensils.

I have been able to find no evidence that any utensils are ever boiled in the hotels.

The following measures should be adopted in order to avoid, or at least materially lessen, the chances of food becoming infected:—

Meat and fish, particularly, should be protected from flies in as effective a manner as possible before it is brought into the hotels.

When in the hotels all food should be protected from flies; the larder should be entirely fly-proof; the entrance should be protected by two doors, between which there is room for a man to stand; both these doors should close automatically with springs, and it would be well to have some simple automatic arrangement which would prevent one being opened until the other was closed. It should be easy to catch the few flies that might possibly get into the larder, in spite of these precautions, by means of fly traps.

Of course the best plan would be to keep the food in a chamber which was constantly below freezing point. When the food was removed, once or perhaps twice during the day, it should be kept in fly-proof receptacles.

Meat should be kept hanging up, and not laid upon shelves.

Shelves and tables in the larder, serving rooms and kitchens should be made of some non-absorptive material, such as marble or slate. Most of the shelves and tables upon which the food was placed during the processes of cooking and serving which I saw in the islands were made of soft wood. No matter how much this wood be scrubbed, there must always be a certain amount of organic material in a more or less advanced stage of decomposition in the cracks. In the serving rooms, kitchens, &c., and wherever food is exposed for any length of time to contagion by flies, the food should be covered up as soon as it is put down. The ordinary wire gauze dish covers are cheap, and admirably suited to this purpose.

Cooking utensils, plates, dishes, forks, spoons, &c., should be sterilized shortly before use. This would not involve any very considerable extra labour, and convenient apparatus would not be very costly. No pressure of steam would be necessary, only the utensils should be brought to the temperature of steam. Cleaning with a jet of live steam, such as is done on ships, would be very effective.

Copper cooking utensils have the disadvantage that they require re-tinning at intervals. There is no means of getting this done in a first-class manner in the islands. The tinning is often irregular, and it is impracticable to get such a surface really clean and free from small collections of organic material. Something other than copper would, therefore, be an advantage.

Soup must be made fresh every day, and the stock-pot abolished.

With regard to rechauffés, even if protection from flies is guaranteed between the first and second cooking, it would be well if the material were always brought to boiling point and kept so for some minutes.

Cold cooked provisions must be kept free from flies.

There should be but little difficulty in keeping the kitchen, and even the whole house, comparatively free from flies by means of wire gauze frames to the windows and double doors: the outside door to consist of a frame with wire gauze stretched upon it. Such a plan would allow plenty of air to come into the rooms, and would exclude the majority of the flies. This is done very extensively in America, and even by some people in England.

No suggestion is intended that the kitchens of the hotels are not

clean in the ordinary acceptance of the word. For instance, the kitchens of those I visited would compare very favourably with any kitchen I have seen in Europe.

What the observations really imply is that precautions which are sufficient in England to prevent a degree of infection by bacteria enough to produce symptoms, are wholly inadequate under the conditions of temperature, &c., in the lower and hotter parts of the islands.

It is quite possible that there may be one or more specific bacteria which are specially responsible for the acuteness of the symptoms. Even if this be the case, however, there seems but little doubt that the flies are to a large extent responsible for the original infection of the food. The rapid multiplication of the bacteria and the consequent production of toxins depends upon the local conditions. It would seem that the suggested precautions are necessary whether there be a specific micro-organism or not. It is probable that food is more frequently infected, even in the best conducted private kitchens in the towns in the islands, than is the case in Europe, and that consequently the residents may have acquired a limited degree of immunity. I met several residents, however, who told me that they had suffered from attacks after dining at hotels, but not at any other time.

The term 'Canary Fever' is obviously a misnomer. It would appear probable that a bacterial infection of the food must frequently happen, and similar epidemics be produced in other tropical and sub-tropical countries where similar conditions prevail.

I wish to acknowledge the great assistance given to me by Dr. Taylor and by Dr. Millares. Their local knowledge was of the greatest value. The enquiry was much facilitated by the ready help and great frankness with regard to the facts that had come within his knowledge, shown to me by Mr. Sauerbrei, the manager of the Hotel Metropole at Las Palmas.

CONTRIBUTION A L'ETUDE DE *POROCEPHALUS MONILIFORMIS*

CONTRIBUTION A L'ETUDE DE *POROCEPHALUS MONILIFORMIS*

PAR

A. BRODEN

DIRECTEUR DU LABORATOIRE

ET

J. RODHAIN

MÉDECIN DE L'HÔPITAL DES NOIRS

Laboratoire de Léopoldville (Etat du Congo)

(*Received November 13th*, 1907)

Nous avons eu récemment l'occasion d'observer à Léopoldville, plusieurs cas d'infection par des larves de *Porocephalus moniliformis*, un cas chez l'homme, quatre cas chez le singe.

Les infections par ce parasite doivent être dans le bassin du Congo comme ailleurs, extrêmement rares. Dans les nombreuses autopsies d'hommes et d'animaux les plus divers que nous avons faites depuis 1900 à Léopoldville, à Lusambo et dans l'Ubangi, nous n'avions jamais rencontré de larves de ce parasite. Dutton et Todd* qui ont fait au Congo également de nombreuses autopsies, n'ont pas signalé de ces larves.

HISTORIQUE

Pour autant que la littérature nous est accessible ici, peu de cas d'infection par *Porocephalus moniliformis* semblent avoir été signalés. Rencontrées une première fois par Pruner en 1846 dans le foie de deux nègres au Caire, des larves furent signalées ensuite par Bilharz, Fenger, Aitken, Giard, Chalmers. Tous ces cas se rapportent à des infections chez l'homme.

Nous donnons cette littérature d'après Braun.[1] Depuis 1899, date de la publication de l'observation de Chalmers, il ne semble pas avoir été signalé de nouveau cas.

* During the autopsies of thirty odd natives and some scores of monkeys (*Cercopithecus sp.*), we saw *Porocephalus sp.* four times; twice in man, twice in monkeys; and in each instance the parasite was firmly encapsuled in the liver. (J. L. Todd).

1 Braun. Thierische Parasiten des Menschen. 3 Aufl. 1903.

D'après les observations de ces différents Auteurs, les larves de *Porocephalus* s'enkystent dans le foie, dans la muqueuse intestinale, dans le poumon. Chalmers[2] en a retrouvé à l'autopsie de nombreux exemplaires libres dans la cavité péritonéale et dans l'intestin grêle.

Dans certains cas, observations Aitken, Chalmers, le parasite semble avoir été la cause directe de la mort par pneumonie ou péritonite.

Chez l'animal les larves de *Porocephalus moniliformis* ont été rarement constatées. Pruner en a signalé chez la girafe, d'autres Auteurs chez l'hyène, le *Cynocephalus mormon* et le *Cercopithecus albogularis* (Looss[3]).

Looss se range à l'avis de Neumann[4] qui considère les larves de *Porocephalus* signalées jusqu'à présent chez l'homme et quelques animaux, comme la forme jeune de *Porocephalus moniliformis*, Diesing. Celle-ci se rencontre dans les poumons des grands serpents africains.

L'évolution du parasite n'est pas connue avec certitude. D'après Looss, elle doit être analogue à celle de *Linguatula rhinaria*.

OBSERVATIONS PERSONNELLES

Le 11.V.07 est amené à l'hôpital des noirs à Léopoldville, le soldat Bangala-Mingi, se plaignant de vives douleurs dans le flanc gauche. Un examen superficiel dénotant un état général grave, le malade est gardé en observation.

De l'interrogatoire et de l'examen faits par l'un de nous, nous retenons les faits suivants :

Le soldat est arrivé à Léopoldville au commencement d'avril 1907 ; avec un détachement il est parti à l'intérieur du district, où il serait devenu souffrant vers le 15 avril. A ses camarades il se serait plaint alors de vagues douleurs dans le flanc gauche et de constipation.

C'est un homme largement bâti, à bonne musculature, à nutrition bien conservée. L'attention est attirée immédiatement par l'abdomen dont la moitié gauche est fortement bombée. A la

[2] Centrlb. f. Bakter. 1899, Bd. XXVI, p. 518.

[3] In Mense, Handb. f. Tropenkrankheiten, Bd. I, p. 198.

[4] Porocéph. du chien et de quelq. mammif. Arch. de Paras. II, 1899. Ce travail ne nous a pas été accessible ici.

palpation l'abdomen présente une résistance uniforme, assez marquée, une résistance anormale dans l'hypochondre gauche et en partie dans l'épigastre. La palpation est très douloureuse en cette région et ne permet pas une délimitation exacte. A la percussion, il y a une sonorité tympanique élevée au niveau de l'hypochondre et du flanc gauches, sans limites nettes. La limite antéro-inférieure de la rate n'est pas décelable ; le foie n'est pas augmenté. Le cœur ne présente pas d'altérations. Le poumon gauche présente un peu d'engorgement à la base, le poumon droit normal.

Pas de trypanosomiase. L'examen du sang révèle augmentation notable des leucocytes (myélocytes). La température est fébrile, 37·8° C., le pouls très accéléré, filant.

Le malade se plaignant de n'avoir pas eu de selles depuis 15 jours, prend un purgatif qui provoque 2 selles.

Dans la nuit du 11 au 12.V.07 le malade a des vomissements sanguinolents dont l'examen ne dénote rien de particulier.

Le malade est très abattu, refuse toute nourriture. Il meurt brusquement dans la nuit du 12 au 13.

Cette sommaire histoire clinique ne nous donne guère d'indications nettes sur la nature de la maladie. Aussi nous nous étions bornés à supposer l'existence d'une tumeur abdominale de nature indéterminée.

Autopsie.—Commencée le 13 à 7 heures du matin. Cadavre d'un homme normalement conformé, dans un état de nutrition bien conservé. En rigidité cadavérique ; pas de lésions cutanées. L'abdomen est fortement ballonné, la peau y est très tendue ; la sonorité à la percussion est d'un tympanisme élevé sur toute la paroi abdominale antérieure.

Après incision de la paroi, il s'échappe de la cavité abdominale des gaz fétides en même temps qu'un liquide épais, sanguinolent, couleur rouge lie-de-vin. La cavité péritonéale renferme environ 200 cc. de ce liquide. A l'examen microscopique on y trouve peu de leucocytes dégénérés, de nombreux bacilles longs, immobiles, isolés, en chaînettes plus ou moins longues ou en filaments.

L'estomac et l'intestin grêle sont fortement distendus par des gaz. Le long de la grande courbure de l'estomac, occupant tout l'hypochondre gauche, il y a une poche à parois assez épaisses, présentant à la face externe de nombreuses tâches hémorrhagiques d'un rouge violacé. Au palper, cette poche ne présente qu'une

résistance molle, plus ou moins fluctuante. Elle est intimement adhérente d'un côté à la grande courbure de l'estomac et à l'extrémité de lobe gauche du foie, en bas à la partie gauche du colon transverse, en haut à la face inférieure du diaphragme, du côté externe à la face interne des côtes, en arrière à la paroi abdominale postérieure.

En examinant les rapports de la poche, avec le gros intestin, nous retournons le grand épiploon, et remarquons à la face postérieure de celui-ci, près du colon transverse, une *larve* non enkystée, vivante, d'un blanc laiteux ; l'extrémité céphalique est largement arrondie, la moitié postérieure s'amincit graduellement et se termine en cône ; le corps présente sur toute son étendue des épaississements annulaires, rappelant à première vue, du moins dans la moitié postérieure, les tours de spire d'une vrille. La larve est légèrement fixée à l'épiploon par son extrémité céphalique ; elle s'en détache facilement et est mise dans de l'eau physiologique où elle continue ses mouvements lents de contraction et d'extension.

A l'incision de la poche, il s'écoule en jet un liquide épais, trouble, sanguinolent, d'un rouge lie-de-vin ; par compression, il en sort comme des caillots de sang nombreux et épais. Incisée largement, la poche est encore remplie à moitié par une bouillie assez épaisse, dans laquelle se reconnaissent des tractus fibreux de la rate. A la paroi de la poche, nous ne trouvons pas d'autre solution de continuité que celle provoquée par le couteau d'autopsie. L'examen microscopique du contenu de la poche montre à côté d'éléments cellulaires plus ou moins bien conservés, de nombreux bacilles longs, identiques à ceux signalés déjà dans l'exsudat abdominal.

Foie: n'est pas hypertrophié, est d'un jaune pâle. Sur la face antéro-supérieure, le feuillet péritonéal viscéral est particulièrement soulevé par des gaz. Dans la pulpe l'examen dénote la présence de nombreux bacilles identiques à ceux déjà signalés.

Estomac: la face externe ne présente pas de lésions macroscopiques. La muqueuse stomacale présente en differents endroits de petites suffusions hémorrhagiques. L'estomac ne présente pas de communications avec la poche.

Oesophage: pas de lésions macroscopiques.

Intestin grêle: uniformément distendu par les gaz, présente à la surface externe, en de nombreux endroits, des tâches hémorrhagiques, petites, irrégulières, d'un rouge vif. Incisé sur toute sa longeur, il ne

présente pas d'ulcérations ni de perforations. Dans la partie supérieure, il renferme de rare *Ankylostomum.*

Gros intestin: présente également à la face externe de petites suffusions hémorrhagiques, irrégulièrement disséminées ; pas d'autres altérations macroscopiques. Dans le contenu du rectum, rares œufs *d'Ankylostomum*, pas d'autres parasites.

Reins, vessie: pas d'altérations macroscopiques.

Cage thoracique: au poumon droit, pas de lésions macroscopiques. Dans la cavité pleurale droite, quelques c.c. d'exsudat, renfermant des bacilles identiques à ceux de l'exsudat abdominal.

Le poumon gauche, par toute sa surface, présente des adhérences fortes, fibrineuses, anciennes, avec la face interne de la paroi thoracique ; la base du poumon adhère fortement au diaphragme, qui à ce niveau présente des suffusions hémorrhagiques. Le lobe inférieure présente à la base en arrière, des noyaux fortement congestionnés.

Cœur: le péricarde renferme environ 50 c.c. d'exsudat sanguinolent, très riche en longs bacilles ; il en est de même du sang du cœur. Les valvules sont fortement hyperémiées, mais ne présentent pas d'autres lésions.

Les *Cultures* sur agar et en bouillon faites avec l'exsudat abdominal et le sang du cœur, ont donné : celles sur agar, 2 colonies de microcoques : celles en bouillon, une culture serrée de coques. Les bacilles longs signalés à l'examen microscopique, appartenaient donc à une espèce anaerobie. Les coques n'ont pas été identifiés. Les bacilles ne se colorent pas par le gram.

CARACTÈRES DE LA LARVE

1. Elle se présente sous forme d'un petit vermicule, d'un blanc laiteux ; le corps est arrondi, composé d'une série d'anneaux, dont le bord postérieur est épaissi et surplombe un peu le segment suivant. L'extrémité céphalique vue par la face dorsale est largement arrondie. A 1·5 mm. environ du bord antérieur se voit un premier sillon peu profond, suivi bientôt d'une série d'autres très nets.

A la face ventrale, l'etrémité céphalique présente à 1 mm. environ du bord antérieur, sur la ligne médiane, un petit orifice circulaire, ou ouverture buccale. Celle-ci ne présente pas d'appendices.

De chaque côté de la bouche, disposés symétriquement, il y a deux

crochets très pointus, d'un jaune d'ambre. Ces crochets, que l'animal sort et rentre altérnativement, sont logés dans une dépression de tegument, où ils sont peu visibles au repos ou après la mort.

A un peu plus de 1 mm. en arrière de l'ouverture buccale, se voit une première indication de sillon.

2. *Corps*: se continue avec l'extrémité céphalique sans différenciation. Il est composé d'une série d'anneaux, les premiers, près de l'extrémité céphalique, étant peu distincts. Ces anneaux ou segments sont au repos, beaucoup plus larges que longs. Vers le bord postérieur ils s'élargissent un peu et s'epaisissent notablement de façon à surplomber la partie antérieure du segment suivant.

La section du corps n'est pas exactement circulaire: la face dorsale est arrondie, mais la face ventrale est aplatie. Dans la moitié postérieure même, la face ventrale présente une surface légèrement concave. Cette concavité est produite par une disposition spéciale du bord postérieur des anneaux à cet endroit. A partir du 10e segment, le bord postérieur à la face ventrale s'amincit un peu de chaque côté de la ligne médiane et au niveau de celle-ci présente une échancrure à concavité dirigée vers la tête. Cette succession d'amincissements et d'échancrures produit une espèce de sillon peu profond.

Le tegument du corps ne présente aucun détail de structure; le bord postérieur des segments ne porte aucune espèce d'appendices.

Le corps est divisé en 18 anneaux plus ou moins nettement distincts, auxquels il faut ajouter le segment portant la bouche et les crochets ou extrémité céphalique et le segment final ou extrémité postérieure. Ce qui ferait 20 segments, mais nous verrons que le dernier anneau présente encore des indications de subdivisions.

3. *Extrémité postérieure:* à l'œil nu se présente sous forme d'un segment conique à pointe obtuse. A un grossissement convenable, on y distingue surtout à la face ventrale, deux épaisissements analogues au bord postérieur des segments du corps, et partageant donc en réalité le segment terminale en 3 petits segments peu distincts. Cela porterait le nombre total des segments à 22.

A la pointe du segment terminal, est un orifice sous forme de fente transversale—orifice d'excrétion (?).

Dimensions de la larve: Durant la vie, les dimensions varient suivant l'état de contraction, ou d'extension de l'animal. La plus

grande longueur que nous ayons mesurée, les trois-quarts environ des anneaux étant en extension, était de 23 mm. Après la mort et fixation par le sublimé, puis alcool à 70%, la longueur est de 16 mm., la largeur de 2 à 2·5 mm., la plus grande largeur correspondant à l'extrémité céphalique, immédiatement en arrière de l'orifice buccale.

La larve extraite à l'autopsie le 13.V.07 dans la matinée put être gardée en vie dans de l'eau physiologique et du sérum du sang de bœuf pendant 6 jours : elle fut trouvée morte le matin le 19.V.07.

Cette larve présente des caractères identiques à ceux decrits par différents Auteurs pour d'autres exemplaires rencontrés chez l'homme, et denommés *Pentastomum constrictum*, von Siebold 1852, *Porocephalus constrictus*, Stiles 1893. Comme nous l'avons dit plus haut, Looss et Neumann considerent ces larves comme la forme jeune de *Porocephalus moniliformis*, Diesing, 1836. Nous adoptons cette dénomination qui nous paraît la plus rationnelle.

Quels ont été les rapports de cette larve de *Porocephalus* avec la maladie et même la mort de notre sujet?

Différents Auteurs ont signalé les ravages causés dans l'organisme par une ou plusieures de ces larves. Les faits constatés jusqu'à présent tendent à prouver que les embryons parvenus de l'une ou de l'autre façon dans les organes, s'y enkystent et s'y développent. Arrivée à un certain stage de développement, la larve romprait la poche et chercherait à sortir de l'organisme dans lequel elle est emprisonnée, pour gagner l'extérieur. Par les crochets dont elle est armée, par sa taille, elle doit nécessairement alors occasionner de graves désordres.

En examinant les lésions macroscopiques constatées à l'autopsie de notre sujet, nous pouvons dire que cet homme à succombé à une *péritonite suraigue consécutive à une fonte purulente de la rate.* Quelle a été la cause des modifications considérables produites dans la rate? Nous ne croyons pas nous tromper en les attribuant à la larve de *Porocephalus*. L'embryon s'est enkysté, s'est développé dans cet organe. Arrivé à un certain stage de développement, la larve a rompu le kyste et a cherché à sortir de l'organe qui l'emprisonnait. Elle a dû produire ainsi une irritation considérable suivie bientôt d'une réaction inflammatoire intense. Ainsi s'expliquent les douleurs accusées par notre malade déjà plus d'un mois avant la mort. Sur ces lésions, s'est greffée une infection microbienne, occasionnant une

véritable fonte purulente de la rate avec un développement considérable de gaz. Peut-être, immédiatement après la perforation de la poche par la larve, s'est développée la péritonite suraigue. Les caractères morphologiques et biologiques du bacille, signalé dans l'abcès, les divers exsudats et le sang du cœur, nous incitent à croire que nous avons eu affaire au bacille de l'oedème malin.

LARVES DE *POROCEPHALUS* CHEZ LE SINGE

Peu de semaines après le cas du nègre que nous venons de relater, nous avons eu la bonne fortune de retrouver de nombreuses larves de *Porocephalus* chez un singe, *Macacus* (*sp.*?).

Ce singe avait été acheté à Léopoldville à un noir descendu du Haut-Congo ; il ne paraissait pas malade et fut gardé en captivité pendant plusieurs semaines. Il fut trouvé mort le matin du 10.VII.07. Bien qu'il n'eût pas servi de sujet d'expériences, nous en fîmes l'autopsie pour déterminer la cause de la mort et contrôler la parasitologie intestinale. A notre grand étonnement, nous trouvâmes de nombreuses larves de *Porocephalus*, 8 enkystées dans le grand épiploon, 1 dans le mésentère, 2 à la face inférieure du diaphragme. Nous en donnons plus loin une description sommaire.

Le 3.IX.07, un autre de nos singes *Macacus* (*sp.*?), de même espèce que le précédent, mourut dans la journée. N'ayant pas servi aux expériences, l'animal fut néanmoins autopsié.

Il présentait dans la cavité abdominale de nombreuses larves de *Porocephalus* : 7 enkystées dans le grand épiploon, 2 dans le mésentère, 1 fixée à la face inférieure du diaphragme, 2 fixées à la paroi du petit bassin.

Pas de lésions macroscopiques des organes.

Le 4.IX.07, un troisième de nos singes *Macacus*, de même espèce que les précédents, succomba. A l'autopsie, nous trouvâmes 13 larves de *Porocephalus* enkystées dans le grand épiploon.

Pas de lésions macroscopiques des organes.

Le 23.IX.07, un 4ième singe, de même espèce que les précédents, fut trouvé mort. Comme chez les autres, le grand épiploon renfermait 12 larves enkystées de *Porocephalus*.

Pas de lésions macroscopiques des organes.

Ce dernier singe avait paru indisposé à certains moments.

Fréquemment on l'avait trouvé étendu, couché sur le ventre, comme s'il souffrait de douleurs abdominales.

Chez ces 4 singes, les larves de *Porocephalus* montraient une disposition identique.

L'immense majorité des larves était fixée au grand épiploon, quelques unes au mésentère, de très rares au péritoine pariétal: pas une seule dans les organes, comme le foie ou la rate.

Comme le montre la photographie faite du grand épiploon du singe No. 3, les larves sont enroulées sur elles-mêmes, entourées d'une mince membrane kystique, à travers laquelle les segments du corps sont visibles. En incisant la membrane près de l'une des extrémités de la larve, une légère pression suffit pour faire sortir le ver de la poche.

La membrane qui enferme la larve, est fibrineuse, très mince, ne montre pas de vascularisation. Le petit kyste qu'elle forme, renferme outre la larve, une très petite quantité de liquide un peu trouble, mais sans éléments cellulaires.

Ce kyste est intimement adhérent à l'épiploon ou au péritoine pariétal, sur lequel il est fixé: on ne peut l'en détacher sans déchirer la séreuse.

Immédiatement autour du kyste, ni l'épiploon, ni le mésentère, ni le péritoine pariétal, ne présentent d'altérations macroscopiques permettant de croire à une réaction cellulaire. Le même dans la cavité abdominale, il n'y avait pas de lésions macroscopiques des organes ni d'exsudat.

L'une ou l'autre des larves, enlevée du kyste et mise dans l'eau physiologique, ont présenté des mouvements analogues à ceux décrits pour la larve du nègre. Elles n'y ont vécu que durant 48 heures au maximum.

Les caractères morphologiques sont identiques à ceux décrits pour la larve du nègre. Alors que celle-ci après fixation au sublimé, a une longueur de 16 mm., une largeur de 2 à 2·5 mm., celles des singes n'ont que 10 à 12 mm. de long, et 2 mm. de large.

Mode d'infection: En présence des cas répétés d'infection par des larves de *Porocephalus* chez des singes tenus en captivité au même endroit, nous devons nous demander si ces animaux ne se sont pas infectés à une source commune.

Ces singes étaient gardés à la chaîne sous la vérandah d'une

habitation, et pouvaient se promener dans un certain rayon sur le sol qui entoure la maison. Ils recevaient comme nourriture de la chikwangue, du pain et du riz cuit, de temps à autre une banane. Tous avaient été achetés, 2, 3 ou 4 mois auparavant à des nègres descendus avec ces animaux du Haut-Congo : il ne fut donc pas possible de connaître plus exactement leur lieu d'origine.

Des quatre singes, seul le 4ième avait de temps à autre montré des signes d'indisposition, en se couchant à plat ventre durant des heures, refusant toute nourriture.

Actuellement nous possédons encore deux singes qui ont été en captivité dans le voisinage immédiat des quatre qui ont succombé et en contact continu avec eux pendant des semaines. Jusqu'à présent ni l'un ni l'autre de ces singes ne présentent des symptômes de maladie.

Si l'on admet pour *Porocephalus moniliformis*, une évolution analogue à celle de *Linguatula*, nos singes auraient dû être infectés par les œufs provenant d'un animal hôte de la forme sexuelle, adulte. Looss, avec Neumann, admet que les grands serpents africains constituent les hôtes des formes adultes. A Léopoldville, les grands serpents sont rélativement rares, et si nos singes ont été infectés pendant qu'ils étaient en captivité chez nous, comme nous sommes portés à le croire, nous serions plutôt tentés d'admettre que les œufs de *Porocephalus* proviendraient d'un autre animal qu'un serpent. D'un autre côté, il ne serait certainement pas impossible que des œufs renfermant un embryon, et très résistants d'après Looss, avaient été véhiculés d'une certaine distance jusqu'à notre laboratoire.

Des circonstances materielles ne nous ont pas permis d'essayer d'infecter d'autres animaux avec les larves provenant des singes.

Dès que l'occasion se présentera, nous examinerons des serpents et d'autres animaux sauvages pour tâcher de retrouver la forme adulte de *Porocephalus moniliformis*.

EXPLICATION DE LA PLANCHE

Fig. 1.—Extrémité antérieure de larve de *Porocephalus moniliformis* du nègre, vue par la face dorsale: Gross. 5 diam.

Fig. 2.—Extrémité antérieure, *id*, vue par la face ventrale.

Fig. 3. Larve de *Porocephalus moniliformis* du nègre, vue par la face ventrale: Photographie en grandeur naturelle.

Fig. 4.—Larve, *id*, vue par la face dorsale: Photographie, *id*.

Fig. 5.—Grand épiploon du singe No. 3, avec larves enkystées: Photographie $\frac{3}{4}$ grandeur naturelle.

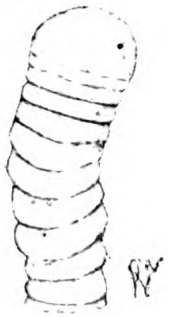


Fig. 1



Fig. 2



Fig. 3



Fig. 4

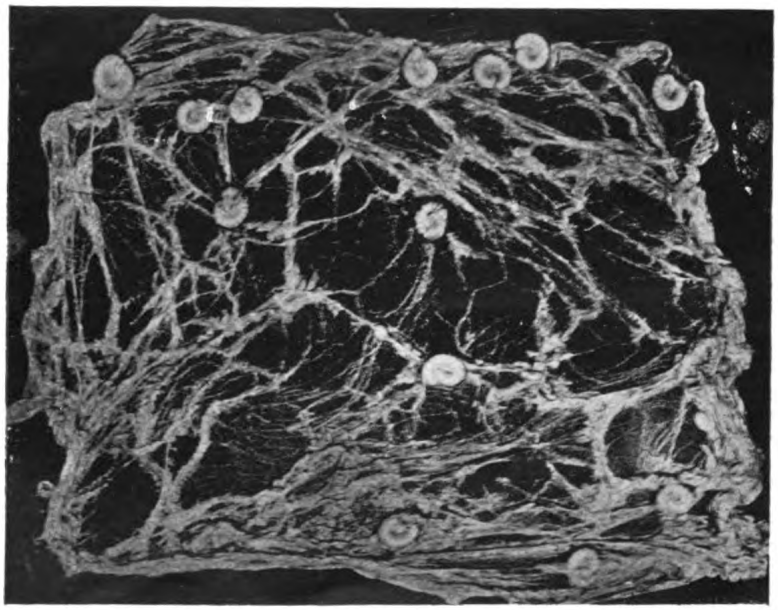


Fig. 5

Braden, phot.

ON THE HABITS, LIFE-CYCLE AND BREEDING PLACES OF THE COMMON HOUSE-FLY (*MUSCA DOMESTICA*, Linn.)

ON THE HABITS, LIFE-CYCLE AND BREEDING PLACES OF THE COMMON HOUSE-FLY (*MUSCA DOMESTICA*, Linn.)

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This paper is reprinted, by kind permission of Dr. E. W. Hope, Medical Officer of Health, from the preliminary report issued by the Health Committee of the City of Liverpool, 3rd October, 1907. Some additional facts regarding the habits of the house-fly have been obtained recently, but it has been thought desirable to embody these in the final report, together with the results of further experiments with insecticides, disinfectants, and other methods of control.

"This investigation was conducted chiefly with the view of ascertaining the nature and extent of the breeding places of the common house-fly (*Musca domestica*) in the City of Liverpool; and also the period of the life cycle of the fly under varying atmospheric and other conditions; so that some practical measures might be devised for the destruction of this pest.

"In addition to the common house fly, other species of flies also occur in dwellings and shops, and several species were bred from ash-pit refuse, human excreta, &c. One of these, the common blow-fly, or blue-bottle (*Calliphora erythrocephala*), may also prove to be a very important contributory factor in the spread of zymotic diseases, but it is intended, for the sake of clearness, to deal with these additional species in the Appendix to this Report.

"In the popular mind the term 'house-fly' is applied to almost all kinds of two-winged flies which are commonly met with in the dwellings of man. To the zoologist, however, there is but one true house-fly, and this is the *Musca domestica* described by Linnaeus in 1758. This fly is by far the commonest species met with, and quite 90 per cent. of the flies which infest houses in Liveprool are of this kind.

"Altogether, the refuse from over three hundred ashpits and bins (chiefly the former) was examined, and 37 middensteads carefully inspected. Human excreta found in the courts and passages were also inspected, and breeding-cage experiments with these and the excreta of domesticated animals were also conducted. The survey was restricted to five areas in different parts of the City embracing, in all, visits to 68 streets.*

"The result of the investigation and survey has proved eminently satisfactory, both from an economic and scientific standpoint. It has led to the discovery of the chief breeding places of the fly, and many new and interesting facts relating to the food of the larval stages have been brought to light; so that we are now in possession of the more important facts relating to the economy of this pest.

"The chief breeding places of the house-fly may be classified under the following heads:-

"1. Middensteads containing horse manure only.

"2. Middensteads containing spent hops.

"3. Ashpits containing fermenting materials.

"Leaving for the present the minor breeding places, we may proceed to consider the chief ones in detail.

"1. Stable middens containing horse manure only, were broadly speaking, found to be the chief breeding places. In the majority of these the larval stages of the house-fly occurred in countless thousands, revelling in the heat produced by fermentation. The adjacent walls often swarmed with newly-hatched flies, and occasionally one also found enormous masses of their eggs (fig. 2), while deep down at the sides, in the cooler portions of the receptacles, the pupa or chrysalis stage occurred in enormous numbers, looking like small heaps or collections of reddish berries. Middens containing a mixture of horse and cow dung were also infected, though to a less extent than those receptacles containing horse manure only. It is important to note, however, that in all cases where fowls (not ducks or geese) were kept and allowed freedom in the yards, relatively few of the earlier stages of the house-fly were found; and whenever present were invariably located in places inaccessible to the fowls. To make certain that the fowls were responsible for so remarkable a diminution of the fly larvae and pupae, a trowel full of these was

* The list of streets and also the nature of the receptacles are here omitted.—R.N.

thrown to some fowls, when they were eaten with as much avidity as if they had been so many grains of wheat. However much, therefore, we may deprecate the keeping of fowls in large towns, we must, from the evidence which has been adduced, consider them as important contributory factors in the destruction of the earlier stages of the house-fly. It should be pointed out, however, that fowls are kept in a very few of the stable yards, so that in the majority of cases the flies go on breeding uninterruptedly, and, so far as one can gather, the larvae and pupae have few, if any, other natural enemies but those already mentioned.

" In one case, where large quantities of a disinfectant (Sharrant's disinfectant powder) were used in the stable, no larvae or pupae were found in the manure, though they were swarming in a mass of waste hops in a separate division of the same midden. Fly larvae were also absent in another instance where chloride of lime had been used freely. However, one is not prepared, at the present moment, to state definitely that the presence of either of these agents had any deleterious effect on the fly larvae, or that they acted as a deterrent; it may have been a simple coincidence, and the matter requires further investigation.

" All types of middensteads were infected—*roofed*, *vaulted* and *open*.

" The photographs (figs. 6-9) submitted with this report will afford some idea of the enormous numbers of the earlier stages of the house-fly which were found in stable manure.

" 2. Only one midden containing warm spent hops was inspected, and this was found to be as badly infested as any of the stable middens. The pupae (fig. 12) were found collected together in large masses, and the larvae swarmed in the warmer parts of the material.

" 3. A great deal of time was given to the inspection of ashpits, and it was found that wherever fermentation had taken place, and artificial heat had been thus produced, such places were infested with house-fly larvae and pupae, often to the same alarming extent as in stable manure. Such ashpits as these almost invariably contained large quantities of old bedding or straw and paper, paper mixed with human excreta or old rags, manure from rabbit hutches, &c., or a mixture of all of these. (See figs. 10, 11.)

" About 25 per cent. of the ashpits examined were thus infested.

"House-flies were also found breeding, in smaller numbers, in ashpits in which no heat had been engendered by fermentation.

"Both open and closed ashpits were infested, but on the whole the flies gave preference to the closed receptacles. On opening the doors of some of the covered ashpits, the flies often came away in hundreds, appearing like bees round a hive. Ashpits which had been previously treated with disinfectants were also infested.

"In addition to the foregoing, there were also other collections of material which afforded temporary breeding places. They are considered under the following heads:—

"1. Collections of fermenting vegetable refuse.

"2. Accumulations of manure at the wharves.

"3. Bedding in poultry pens.

"1. Collections of straw mixed with other vegetable matter and feathers, lying in open spaces in poultry yards, were found to contain enormous numbers of house-fly pupae (fig. 13). The materials had evidently fermented, and had also been lying exposed to the weather for a period of not less than eight weeks.

"2. The large accumulation of stable manure lying at the wharf (Carruthers Street) was swarming with fly larvae, and the flies occurred also in large numbers; so that it is quite evident that a number of the insects hatch before the manure is placed in the barges for transmission to the country. Practically all the manure is badly infested when it reaches the wharves, so that it is important that it should be shipped with as little delay as possible.

"3. The common practice of leaving 'bedding' material (chiefly long straw) in poultry pens (containing ducks, geese, &c.) until fermentation takes place, affords breeding places for house-flies, and such material should be removed at much shorter intervals.

"Temporary breeding places destroyed by speedy removal of refuse:—

"1. Collections of stable manure removed at short intervals.

"2. Ashpits emptied at intervals of 7-10 days.

"3. Bedding in piggeries.

"1. Ashpits emptied at intervals of about 14 days may be considered as temporary breeding places, the period between the removals being too short for the fly to complete its life cycle. Only under very unusual circumstances, where, for instance, a large amount

of fermenting materials are thrown into the receptacle immediately after it has been emptied, would it be possible for the insect to complete its metamorphosis in so short a period. In the majority of cases, therefore, ashpits emptied at such short intervals can only be considered as temporary breeding places.

" 2. Larvae of the house-fly occurred in large numbers in the receptacle used for holding stable manure in the Corporation Yard,* Mill Lane. As the manure is removed from this place every week, it is impossible for the flies to complete their metamorphosis in it.

" 3. 'Bedding' in the 'piggeries' which had been in use for a few days was found infested with fly larvae. The material used as 'litter' was wood chips. This refuse does not lie sufficiently long for the insect to complete its metamorphosis.

" The non-breeding places were :—

" 1. Middens containing excessively moist and non-fermented cow-dung.

" 2. Patent ash bins.

" 3. Refuse in ashpits saturated with water.

" 4. Human excreta lying in courts and passages.

" 1. No trace of house-fly larvae or pupae was found in any of the middens containing cow manure exclusively. The excessive amount of moisture present in this material was evidently unfavourable to the development of the larvae. But in all cases where horse manure was mixed with it, and fermentation had taken place, larvae were present. The admixture of straw with cow manure would produce similar results.

" 2. No house-flies were found breeding in the patent bins, which is due to the fact that they are emptied daily, or at intervals of a few days.

" 3. No house-flies were found breeding in excessively dry refuse, or refuse thoroughly saturated with water and reeking with inky black putrid matter.

" 4. Human faecal matter found lying in the courts and passages was not infected with fly larvae, and no house-flies were bred from such matter. But human excreta found in ashpits (several instances) and stable middens (two cases) were found to be infested.

* It is interesting to note that the bedding material used at this place was sawdust.

"Reference has already been made to the more important materials in which the larvae were found breeding, but no details have been given as to the exact nature of their food. The dietary, so far as one has been able to see, is almost exclusively that of moist decaying vegetable matter. Horse manure and spent hops were most favoured; but they revelled also in rotten flock beds, straw mattresses; they thrived on old cotton garments and rotten sacks, and waste paper; and seemed especially partial to the dirty beddings from rabbits and guinea pigs. They fed also on bread, decayed fruits and vegetables, the excreta of domestic fowls and pigs, and on human excreta in the ashpits and stable middens. In all cases, however, they thrived best and occurred in the greatest profusion where fermentation had taken place, so that the temperature of the habitat was raised above that of the outside air.

"Like nearly all its congeners, the house-fly undergoes a complete metamorphosis, in which there are *four* well-marked stages; these stages in the life cycle are:—

"1. The egg (ovum), commonly known as 'fly-blows.'
"2. The larva or maggot stage.
"3. The pupa or chrysalis stage.
"4. The imago, or perfect fly. (The final stage.)

"1st Stage.—*The eggs* are laid in small irregular clusters, or in large collective masses* (figs. 1, 2) consisting of many thousands of individual eggs. They are almost invariably deposited on or in such substances as will provide food for the larvae or maggots. They are usually placed in narrow crevices near the surface, but, occasionally, also at a distance of from four to six inches below the surface, the *favoured spots* in all cases being fermenting vegetable matter or the refuse lying immediately over such materials, or in refuse that is likely to ferment. They are often laid, however, on materials which do not ferment, and in all such cases (in this country, at least) the developmental cycle is greatly prolonged.

"The eggs are pure white, and present a highly polished surface, due to the clear, viscous substance with which they are coated. The form of the egg may be seen on referring to the micro-photographs

* I have witnessed as many as 78 females laying their eggs in one small area, the flies being closely packed together. When disturbed they flew away, but returned again to the same spot in a few minutes.—R. N.

(figs. 3, 4), and the actual size is shown in fig. 1. The larvae or maggots hatch from these in periods varying from eight hours to three to four days; the average time may be given as twelve hours, but when laid in fermenting materials the incubation period is reduced to a minimum of eight to twelve hours.

"The number of eggs laid by a single fly averages from 120 to 140. More than one batch may be laid during the life of the fly, but this question has not been definitely settled.

"2nd Stage.—*The larva or maggot* (fig. 9) resembles that of the common 'bluebottle-fly,' or 'Blow-fly' (*Calliphora erythrocephala*), but is much smaller, measuring when fully matured nearly half an inch in length, and is distinguished also by certain anatomical characters. It is essentially a vegetable feeder; animal matter is eaten only, so far as one has been able to gather, when in the form of human faeces. It was never found feeding on the carcases of dead cats and dogs or of birds and fish remains.*

"They thrive and mature most rapidly, and are always most abundant in fermenting materials; but they can also mature in non-fermenting substances during warm weather, though under such conditions they do so very slowly. In stable manure they are generally most numerous a few inches below the surface, and undoubtedly work their way upwards day by day into the fresh material, a few hours (five to six) after it has been added to the previous accumulation. This marked habit is evidently due to the excessive heat which is engendered in the lower strata of the manure.

"Under the most favourable conditions as to temperature and food supply they mature in five to eight days; but, where fermentation does not take place, this stage, even in hot weather, may be prolonged to several weeks (six to eight). In middensteads the fully matured larvae crawl away to the sides or to the top of the wall or framework of the receptacle; in ashpits they locate themselves in various materials as well as ashes, but are evidently partial to old bedding, paper and rags, usually in or near the centre of fermentation. After emptying the alimentary tract of organic matter, pupation takes place rapidly, and the third stage is reached.

"3rd Stage.—The pupa or chrysalis (figs. 7, 10, 11) is at first of a pale yellowish colour, but rapidly changes to bright red, and finally to a dark chestnut colour. It is somewhat barrel-shaped, and varies

* Taschenberg in 1880, states that he found eggs on the carcases of dead animals, but does not indicate that the larvae will breed in such substance.—R. N.

in length from $\frac{3}{12}$ - $\frac{5}{12}$ of an inch. Small examples are found where the temperature has been low or excessively hot and somewhat dry. Large examples invariably occur in fermented materials, more especially so in stable manure.

" In stable middens the pupae occur chiefly at the sides or at the top of the wall or framework of the receptacle, where the temperature is lowest. In such situations they were often found packed together in large masses numbering many hundreds (see figs. 6, 7). The flies emerge from the pupae, under the most favourable conditions, in five to seven days. In ashpits they occur in the positions already indicated, and if similar conditions as to heat prevail, the period is approximately the same; but in all cases where heat is not produced by fermentation, the pupal stage may last from 14 to 28 days, or even considerably more.

" 4th Stage.--The perfect fly escapes from the pupa by breaking away the anterior end; this it accomplishes by inflating the frontal sac, which is situate in the front portion of the head, between the eyes. By the inflation of this sac, the fly is also enabled to force its way through the manure or ashes into the open air. When once it has liberated itself, the wings develop, and when the integument has sufficiently hardened the fly takes to wing. Pairing then takes place, eggs are laid, and another generation is started. The whole cycle from egg to perfect insect occupies, under the most favourable conditions, from ten to fourteen days; but in low temperatures the whole cycle may extend to several weeks. No growth takes place after the wings are developed.

" That the flies migrate from their breeding places to man's dwellings is quite evident, even to the layman, the primary object being apparently to obtain food and shelter. Many flies are also found in such places during winter and early spring, though whether they pass the winter entirely in this stage one has not yet been able to ascertain. It is highly probable, however, that some pupae* may remain over the winter and hatch in the following spring. Now that the breeding places have been discovered, it will be a comparatively easy matter to ascertain if this is the case or not.

* A single pupa evidently of the house-fly was found during the winter months, but this unfortunately did not produce a fly in the following year.

"Little need be said as to the food of the fly itself, as every layman is conversant with the feeding habits of this little filth-carrying insect. But the contributory part which many authorities have claimed that this insect plays in the transmission of zymotic diseases is due to the almost persistent habit it has of feeding or alighting upon human excreta.

"In the course of my investigations, more especially on hot days, numbers of house-flies were seen hovering over or feeding upon such matter. The faeces were generally those of children, and were lying, as a rule, a few feet from the doorways in the courts or in the passages behind the houses. In one instance no less than five patches of human excreta were lying in one court, and all of these were attended by house-flies.

"Temperature, as has already been stated, has a most marked effect upon the developmental cycle of the fly; and a sudden check from heat to cold will materially prolong any one of the stages. Eggs hatched in eight to twelve hours in a temperature of from 75° to 80° F., at a temperature of 60° F. in twelve hours, but at 45° F. they did not hatch until the third day, and then only when placed in a warmer temperature for the purpose of studying them under the microscope. The larvae or maggots mature in the shortest period in fermenting materials at a temperature of between 90° and 98° F., but they usually leave the hotter portions of the stable manure when it reaches a temperature of 100° to 110°. At 54° F. both larval and pupal stages are considerably prolonged; larvae kept at this temperature had not matured at the end of eight weeks, and a number of pupae kept under similar conditions did not produce flies until the fourth and fifth weeks.

"In this Report I have endeavoured to show that:—

"I.—The chief breeding places of the house-fly are:—

"(A) Stable middens containing fermenting horse manure or a mixture of this and cow dung;

"(B) Middens containing fermenting spent hops; and

"(C) Ashpits containing fermenting vegetable matter, or about 25 per cent. of the total number of pits examined.

"II.—That covered ashpits and middens were as badly infested as those which were open.

"III.—That house-flies breed in all temporary collections of fermenting matter.

"IV.—That house-flies breed in relatively small numbers in ashpits where no fermentation takes place.

"V.—That they do not breed in ashpits which are *emptied at short intervals*, or in the *patent bins*.

"VI. That the use of disinfectants in ashpits does not prevent the flies breeding in such receptacles.

"VII. That very dry or excessively wet ashes or moist cow dung* does not harbour them.

"VIII.—That the presence of fowls (not ducks or geese) which had free access to the stable middens reduced the number of larvae and pupae to a very marked extent.

"IX.—That the life cycle of the fly, in all kinds of fermenting materials, is reduced to the minimum period of ten to fourteen days; and that in the absence of such artificial heat the cycle may occupy a period of from three to five weeks or more, according to the temperature of the outside air.

"X.—That house-flies do not depend entirely upon excessively warm weather for breeding purposes, though in hot seasons they would breed much more rapidly in non-fermenting materials, and their numbers, under such conditions, would be greatly increased.

"If house-flies are to be reduced to a minimum, I would submit the following suggestions

"1. That stable manure and spent hops should not be allowed to accumulate in the middensteads during the months of May to October inclusive, for *a period of more than seven days*.

"2. All middensteads should be thoroughly emptied and carefully swept at the period stated in 1.

"The present system of *partly emptying* such receptacles should in all cases be discontinued.

"The walls of middensteads should also be cemented over, or, failing this, the brickwork should be sound and well pointed.

* In excessively hot summers cow dung may form a breeding place for the house fly. The admixture of a large quantity of bedding (straw or sawdust), would also render it suitable for breeding purposes.

" 3. That all ashpits should be emptied, during the summer months, at intervals of not more than ten days.

" 4. That the most strenuous efforts should be made to prevent children defaecating in the courts and passages; or that the parents should be compelled to remove such matter immediately; and defaecation in stable middens should be strictly forbidden. The danger lies in the overwhelming attraction which such faecal matter has for house-flies, which latter may afterwards come into direct contact with man or his foodstuffs. They may as Vedeer* puts it, 'in a very few minutes . . . load themselves with dejections 'from a typhoid or dysenteric patient, not as yet sick enough to be 'in hospital or under observation, and carry tne poison so taken up 'into the very midst of the food and water ready for use at the next 'meal. There is no long roundabout process involved.'

" 5. Ashpit refuse, which in any way tends to fermentation, such as bedding, straw, old rags, paper, waste vegetables, dirty bedding from the 'hutches' of pet animals, &c., should, if possible, be disposed of by the tenants, preferably by incineration, or be placed in a separate receptacle so that no fermentation could take place. If such precautions were adopted by householders, relatively few house-flies would breed in the ashpits, and the present system of emptying such places at longer intervals than, say, four to six weeks, might be continued.

" 6. The application of Paris Green† (poison) at the rate of 2 ozs. to one gallon of water to either stable manure or ashpit refuse will destroy 99 per cent. of the larvae. Possibly a smaller percentage of Paris Green might be employed with equally good results.

" One per cent. of crude atoxyl in water kills 100 per cent. of fly larvae.

" The application of either of these substances might, however, lead to serious complications, and it is very doubtful whether they could be employed with safety. Paris Green, at the rate of 1 to 2 ozs. to 20 gallons of water, is used largely as an insecticide for fruit pests. It does no harm to vegetation when applied in small quantities; but cattle might be tempted to eat the dirty straw in manure which had

* M. A. Vedeer, M.B. 'Flies as spreaders of sickness in camps.' Medical Record, Vol. LIV (1898), pp. 429-430.

† This substance is a definite chemical compound of arsenic, copper and acetic acid.

been treated with this substance, and the results might prove fatal if large quantities were eaten.

"7. The use of sun-blinds in all shops containing food which attracts flies would, in my opinion, largely reduce the number of flies in such places during hot weather. Small fruiterers' and confectioners' shops, as a rule, are not shaded by sun-blinds, and in their absence flies literally swarm on the articles exposed for sale.

"8. The screening of middensteads with fine wire gauze would, undoubtedly, prevent flies from gaining access to manure, &c., but it is very doubtful if this method would meet with any marked success. The gauze would rapidly oxidise, the framework supporting it would probably warp, and numbers of flies would be admitted whenever the receptacle was opened. Moreover, the erection of such a structure would prove a great inconvenience and a hindrance to the removal of the refuse. This, however, does not prejudice the possibility of erecting a good fly-proof screen in the future.

"Experiments with crude carbolic acid, cresylic acid, &c., are being conducted, and the results will be reported later.

"In the introductory remarks on the house-fly, reference has been made to other flies which were found frequenting houses, or were bred from refuse and excreta during the course of investigation. The following is a list of the insects, together with short notes on their habits and prevalence:—

"1. *Calliphora erythrocephala.* The 'Blow-fly.'

"In some parts of Liverpool this fly is quite as abundant as the house-fly, and, like the latter, may, from its disgusting habit of feeding upon faecal matter, also act as a contributory agent in the spread of zymotic diseases.

"This fly feeds upon the faeces of man to a greater extent than the house-fly; moreover, it is also partial to fruits of various kinds, especially over-ripe plums and grapes and dried figs and dates, so that one can readily conceive how it would be possible for the flies to mechanically transmit the germs of disease from faecal matter to fruit, and as a large percentage is eaten uncooked, the transmission of infected faecal matter, if present, would be direct.

"The female insect lays its eggs in all kinds of raw and cooked meat, the carcases of mammals, birds, fish, &c., and wherever such remains were found in ashpit refuse, the larvae of this insect swarmed.

" 2. *Scatophaga stercoraria.* ' Dun ' or ' Yellow Cow Fly.'

" This is a rather large yellow fly which abounds in the country, but is relatively scarce in the city of Liverpool. It breeds in cow dung, and was occasionally seen in some of the middens. It rarely enters houses, and is therefore of no economic importance.

" 3. *Borborus equinus.*

" A minute fly which literally swarms in stable manure all over the city. As it rarely enters houses or shops, it is of no economic importance. Fowls eat large numbers. It breeds in the faeces of the horse.

" 4. *Stomoxys calcitrans.* ' The Stable Fly.'

" This is a blood-sucking insect, and is one of the recognised carriers of Trypanosomiasis in tropical countries. In 1906 it was common in some parts of Liverpool, but this year it has been quite scarce. It breeds in fermenting horse manure and grass mowings. It sometimes enters houses and bites both man and his domesticated animals. Of little economic importance in Liverpool.

" 5. *Homalomyia canicularis.*

" This species is often common in the dwellings of man, and is for this reason often mistaken for the house-fly. In Liverpool, however, it is by no means abundant, and not more than one per cent. of the flies captured in fly-traps are of this species. It breeds in horse manure, and possibly also in the faeces of other animals.

" 6. *Anthomyia radicum.* ' Root Fly ' or ' Root Maggot.'

" Closely related to the foregoing species, but not common in Liverpool. A few examples were captured in fly-traps; but no specimens were bred from refuse of any kind.

" 7. *Homalomyia scalaris.*

" The larvae of this species were frequently seen in ashpit refuse; and a number of these flies were bred from human faeces. It is not a very abundant species, however, and so far as my investigations have gone, it has not been found very often in houses or shops. The larvae feed on all kinds of faecal matter, and are especially partial to human excreta; they revel in privies, often congregating together on the shield board in hundreds. Cases of intestinal myiasis in man have been attributed to the larvae of this fly.

"8. *Psychoda (?) phalænoides.* 'Owl Midge' or 'Moth Fly.'

"A minute moth-like insect often seen on window-panes in houses. The larvae of this insect were common in human faeces, and many examples of the flies were bred from this material. It is also common in putrid sewage matter. May be looked upon more in the light of a scavenger, but is of little or no economic importance.

"9. *Coleoptera.* (Beetles.)

"Two species were particularly common in ashpits, viz., *Creophilus maxillosus* and *Philonthus politus.* They act as scavengers, feeding upon all kinds of débris. They rarely enter houses."



FIG. 1. FOUR BATCHES OF EGGS.
NATURAL SIZE

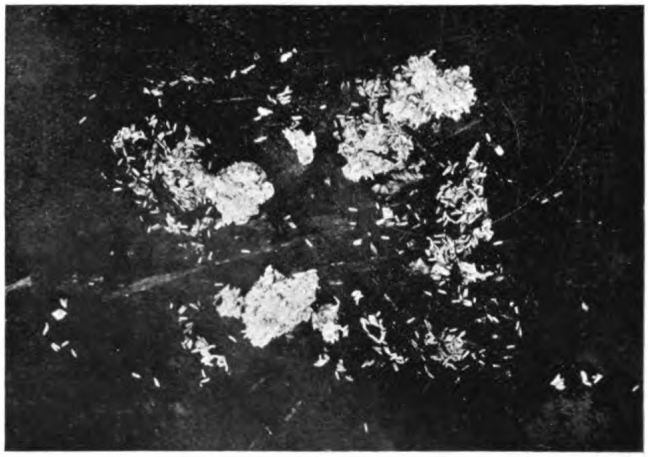


FIG. 2. COLLECTIVE BATCHES OF EGGS IN STABLE MANURE, NUMBERING ABOUT 1,500. NATURAL SIZE



FIG. 3. EGGS ENLARGED

R. Newstead—Photo.

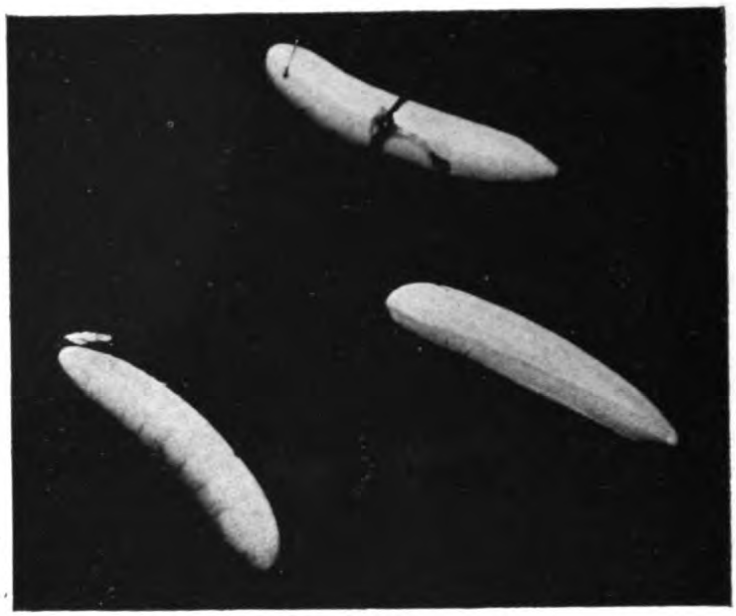


FIG. 4. EGGS GREATLY ENLARGED; ONE SHOWS THE SEGMENTS OF THE LARVA THROUGH THE CUTICLE

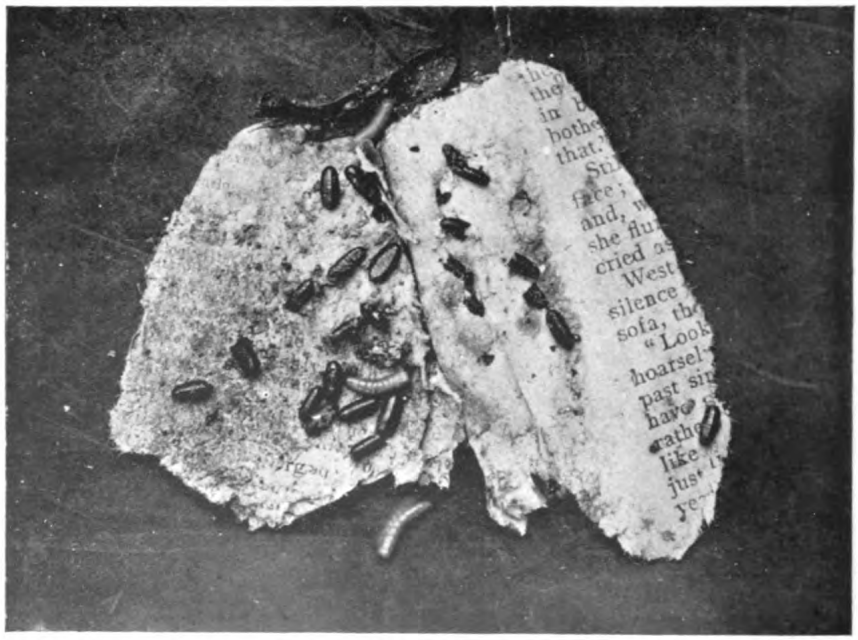


FIG. 5. LARVÆ AND PUPÆ IN WASTE PAPER (ASH-PIT REFUSE). NATURAL SIZE

R. Newstead—Photo.



FIG. 6. MASS OF ABOUT 200 PUPÆ IN STABLE MANURE.
NATURAL SIZE



FIG. 7. MASS OF PUPÆ SEPARATED FROM STABLE MANURE SHOWN IN FIG. 6.
NATURAL SIZE

R Newstead—Photo.



FIG. 8. MASS OF LARVÆ IN STABLE MANURE. NATURAL SIZE

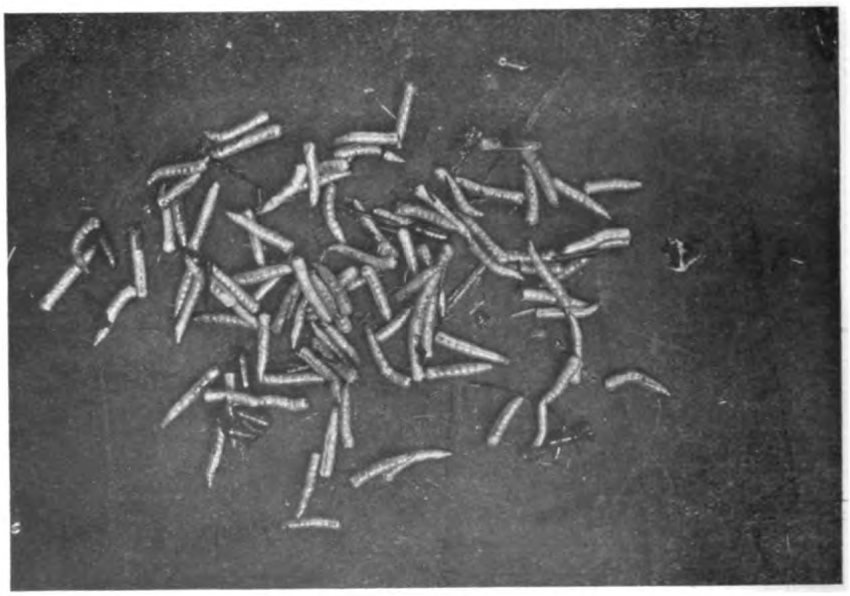


FIG. 9. MASS OF LARVÆ SEPARATED FROM STABLE MANURE SHOWN IN FIG. 8. NATURAL SIZE

R. Newstead—Photo

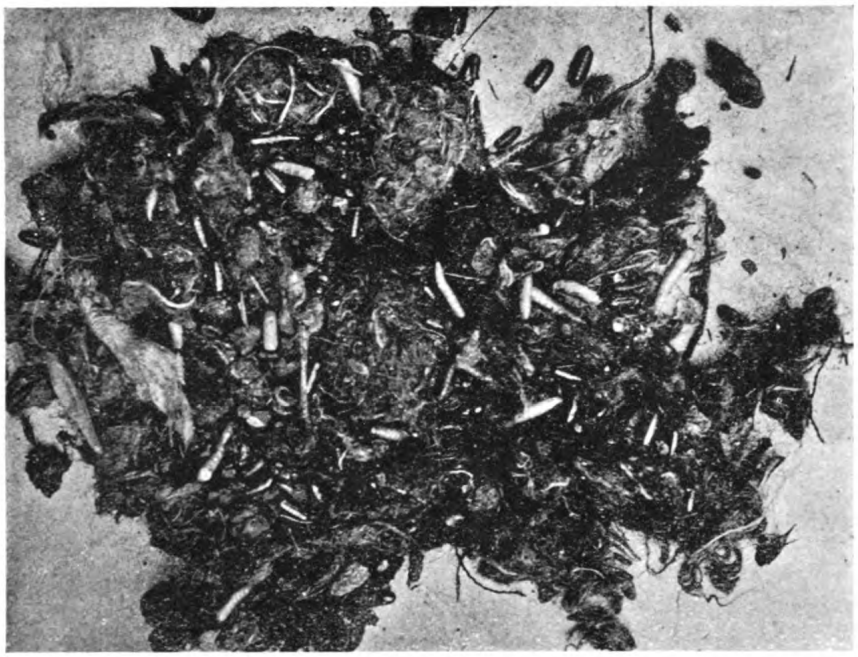


FIG. 10. LARVÆ AND PUPÆ IN OLD "FLOCK" BEDDING.
NATURAL SIZE



FIG. 11. LARVÆ AND PUPÆ IN OLD RAGS (ASHPIT REFUSE).
NATURAL SIZE

R Newstead—Photo.



FIG. 12. MASS OF PUPÆ IN SPENT HOPS. NATURAL SIZE



FIG. 13. PUPÆ IN FEATHERS AND STRAW, FROM POULTRY YARDS. NATURAL SIZE

R. Newstead—Photo.

SOME NOTES ON THE MORPHOLOGY OF *SPIROCHÆTA DUTTONI* IN THE ORGANS OF RATS

SOME NOTES ON THE MORPHOLOGY OF *SPIROCHÆTA DUTTONI* IN THE ORGANS OF RATS

BY

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The strain of *Spirochæta duttoni*, brought from the Congo by Dutton and Todd, is maintained in the Runcorn Research Laboratories through inoculation of infected blood from rat to rat. By Levaditi's[1] silver method I have investigated the morphology of the parasites in the organs of some of these animals killed at different stages of the infection.

The principle of the silver method may be briefly stated as follows:—The organs, after fixation in formol, are impregnated with a solution of silver nitrate, and then exposed to the reducing action of pyrogallic acid. In thin paraffin sections the tissues appear of a bright yellow colour, with sufficient differentiation of the nuclei; the spirochaetes black, with sharply defined outlines. To bring out the relation of the parasites to the tissues, Giemsa's or other stain may be employed.

The pyridin-silver modification, the 'second method' of Levaditi, gave generally good results, not only with fresh material, but also in organs which had been kept for some months in formol.

Details of the course of experimental spirochaete-infection in rats may be found in the extended study of Breinl and Kinghorn;[2] here it is only necessary to repeat that the maximum number of parasites in the peripheral circulation is reached on about the fifth day after inoculation, after which the spirochaetes disappear rapidly and completely, until the first relapse, which occurs in about a week and extends over one to three days.

Examination of sections (4 to 6μ) of the organs of rats, killed at the height of the infection, shows the large and small blood-vessels and capillaries crowded with spirochaetes, in accordance with the observations of Levaditi.

The outlines of the parasites are best seen in the capillaries of the lung and liver, whereas in sections of the heart and large blood-vessels the spirochaetes are generally found massed in broad strands. In the spleen at this stage a different condition is observed; the parasites being comparatively few in number, and mostly intracellular.

The shape of the spirochaetes is not everywhere the same; characteristic organisms, showing regular spirals, being generally seen in lung and heart; while the liver and the spleen contain atypical forms, which can be recognised only by their staining reaction, and by the conditions under which they are found and by the presence of intermediate stages. These forms are seen as small, circular or oval, tightly-coiled spirals, about half the size of a red blood-cell.

Similar forms have been described and figured by Breinl and Kinghorn. These authors found "occasionally in films made from the liver and the spleen spirochaetes coiled up into a small compass, staining a deep red with Giemsa's stain, and surrounded by a well-stained membrane." Levaditi has also observed them as occurring in the liver of mice, and discusses their probable nature in the light of similar appearances met with by himself, Manouélian, Cantazucène, v. Prowazek,[3] Schaudinn, and other authors, in the spirillosis of fowls. He interprets them as agony-forms, preceding further stages of degeneration and disintegration. v. Prowazek, on the contrary, considers these bodies as "resting stages" in the life cycle of the parasite.

With regard to this interesting question, I may state that these forms occur in by far the greatest numbers in the liver, less numerously in the spleen, scantily in the lung, while in sections of the heart-blood they could not be demonstrated with certainty.

Moreover, very few, if any, have been observed free in the lumen of the capillaries, the majority being easily demonstrated in phagocytes.

These two facts—the excessive disproportion in the number of these altered forms found in the liver, and their approximately constant relation to phagocytes—seem to afford a strong argument in favour of Levaditi's hypothesis.

Observations were also made at the crisis of the disease. Breinl and Kinghorn have already pointed out the difficulty of determining this period. In one case described by these authors, the disappearance

of the parasites from the peripheral circulation occurred between 2 and 5-30 a.m.

In the present case, material at this period was obtained with less difficulty. A rat (1,540 B) inoculated on July 20, 1907, showed very numerous parasites in thick films of tail-blood on the morning of July 25. It was examined five hours later, when the parasites were found to have disappeared almost entirely from the peripheral circulation; the animal was then killed for the purpose of examining the organs at the stage of crisis.

Very few spirochaetes were observed in the vessels and capillaries of lung and kidney. The spleen showed a moderate number of large phagocytes containing partly digested and fragmented spirochaetes. In the liver, on the contrary, the capillaries were crowded with parasites, or, to be more exact, were nearly or quite occluded by swollen mononuclear phagocytes—endothelial cells and cells of Kupfer—filled with innumerable black granules, many coiled forms and only a few normal spirochaetes.

This observation seems to lend additional support to the hypothesis that the coiled forms are due to the influence of phagocytosis. At the height of the infection the vessels of the different organs were crowded with free, apparently unaltered spirochaetes, and the liver showed in addition a great number of intracellular coiled forms; at the crisis the extracellular parasites had disappeared almost entirely, and in the liver were found, besides the cell-included coiled forms, large numbers of fragmented parasites and granules, evidently products of intracellular digestion.

The occlusion of many of the capillaries of the liver by swollen phagocytes seems to account for the hemorrhagic and anaemic infarcts observed by Breinl and Kinghorn. Levaditi is also inclined to explain the changes observed by him in the liver of infected mice, by obstruction of the blood-vessels of this organ.

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MALARIA AND HISTORY

MALARIA AND HISTORY

BY

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It seems likely that disease has exercised considerable influence upon the history of mankind. In the struggle for existence, man, by his intelligence, has long since removed wild beasts from the number of his competitors. In civilised parts of the world, at least, the struggle is now limited to competition with his fellow-men and with the parasites of disease. The study of man's combat with man is history as at present understood; the antagonism of man and parasites may prove to be equally important. Although the biological study of disease is still in its infancy, the time has come to collect material, and by careful induction to try to discover any laws which may appear to have a temporary validity. Furthermore, it is well so to limit the investigation as to explore more thoroughly a narrower field before attempting to draw wider inferences or to formulate more general laws.

The effects of endemic disease are easier to investigate than those of epidemics, because the influence exerted is continuous, and spread over a long period. For a similar reason the question is better studied from the historical side than by a consideration of the present state of countries where disease is endemic; although, of course, the latter method will furnish valuable information of which use must be made. Among endemic diseases, malaria, from its wide extension, the large percentage of a people attacked by it, and its long history, appears to afford the best starting-point for the enquirer.

Convinced of the truth of the above statements, the writer began to enquire whether malaria played any part in the history of ancient Greece and Rome, the decline of which is generally thought to have commenced during the fourth century B.C. in the case of the former, and in the second century B.C. in the case of the latter. The suggestion had already been made by Major R. Ross, but there was

practically no previous literature on the subject, as the valuable paper by Professor Aristotle Kouzis, *τινὰ περὶ ἐλειογενῶν πυρετῶν*, Athens, 1907, had not yet appeared. On the other hand, the ancient literature containing references to malaria was of immense size, and none of it could safely be neglected. The means used to identify malaria in ancient literature were as follows:

(1) Tertian and quartan fevers are almost certainly malaria.
(2) Quotidian fevers are very likely malaria.
(3) Enlargement of the spleen and early autumnal fevers very often mean malaria.
(4) Fever in marshy districts is probably malaria.

Of course the ancients knew nothing about microscopes and the action of quinine.

MALARIA IN GREECE

There is an early reference to *πυρετὸς* in Homer, but the word means there, in all probability, "heat" not "fever." The only other possible reference in early times is in Theognis, who lived about 540 B.C. at Megara. He talks in one passage of *ἠπίαλος*, which certainly in later Greek often means ague. In a recent volume, however, of Pauly-Wissowa's Classical Encyclopaedia, it is suggested that this word was originally connected with the nightmare demon. In that case Theognis may be referring to the fright-rigors of nightmare, and not to malaria.

It is important to notice that Hesiod, the Boeotian poet, does not mention malaria as one of the farmer's plagues, though we can be certain that he would have done so had the disease existed. Boeotia is now highly malarious, but in very early times just those places in it were chosen for habitation which are most unhealthy at the present day. Surely malaria cannot have been in this district from prehistoric times. It is true that Suidas quotes *ἐπιάλτης* from some lost work of Hesiod, but although this word is said to mean ague in later Greek, it certainly meant nightmare at first.

In the Hippocratic writings, which may be placed approximately between 450 and 350 B.C., all the "tests" of malaria given above are mentioned again and again, and it must not be forgotten that these writings certainly imply a long tradition behind them. Accordingly, in the Eastern parts of Greece, the home of the doctors who wrote

these traditions, malaria was probably known as early as 500 B.C. If so, it may have come from the East, and malaria is frequently mentioned in the Sanscrit writings. (See Jolly, *Grundriss der Indo-Arischen Philologie und Altertumskunde, medicin*, p. 72.)

But, if the literary evidence can be trusted, the disease did not appear in Attica before 450 B.C., became more common during the period 430-400, and finally was so wide-spread as to be designated in the common speech, though not in the medical writings, by the words πυρετός, πυρέσσω, without further qualification. Every medical writer after Hippocrates mentions malaria in the clearest possible language. Even the remittent forms are discussed, although at first the Greek physicians seem to have been often unable to distinguish remittent from continuous fevers—a fact not to be wondered at when we remember that thermometers did not yet exist. Double and mixed infections were recognised after a while, and Major Ross believes that in the "semi-tertian" we are to see the double malignant tertian. Malarial cachexia, with its attendant evils, splenomegaly, anaemia and dropsy, is described again and again, usually in connection with marshy places. "Fever" is often ascribed to over-fatigue, and it is well known that in a highly malarious country exertion will usually precipitate an attack. Typhoid, clear descriptions of which are curiously lacking in the ancient medical writings, generally takes in them a malarial form, exhibiting tertian periodicity without the peculiar characteristics of typhoid, diarrhoea and rose-coloured spots, while pain in the region of the liver is far more common than in ordinary forms of the disease. Now typhoid often assumes this form in malarious districts, as the doctors of the non-malarious North discovered when, in the American Civil War, they treated patients in the malarious South.

In the non-medical writers malaria is mentioned many times, though not so often as in Latin literature. I can find clear references in Sophocles, Aristophanes, Plato, Aristotle, Demosthenes and the inscriptions.

It is quite impossible, in the absence of direct testimony, to say when malaria first came into Greece. But the fact that πυρετός in the sense of "fever" does not occur before the time of Hippocrates, combined with the probability that at first older people were frequently attacked, makes it likely that the disease did not become

endemic, at least in Attica, until the close of the fifth century B.C. But although we cannot date its introduction, it is quite certain that it became more common after 400 B.C., until a Greek could talk of his touch of fever in much the same way as we talk of influenza, or an ordinary cold. Nor did malaria change its type. From Hippocrates to Joannes, from the fifth century B.C. to the fourteenth century A.D., malaria is described in almost precisely the same terms, and with exactly the same symptoms, and these symptoms are so clear that we can recognise all the various forms of the disease as we meet them at the present time.

MALARIA IN ITALY

When and how malaria was introduced into Italy, or when it became endemic there, is a very complicated question. It is generally assumed, e.g. by Celli, that malaria was common in pre-historic times, and that the drainage works at Rome were intended to diminish the disease, while Cicero is quoted as putting on record that Romulus founded a city in a healthy spot surrounded by an unhealthy neighbourhood. It is quite possible that even in early times the disease did exist, but if this be so, it cannot have been severe, nor can it have been widely spread. For it is certain that some districts, the most highly malarious in the historical period, were flourishing centres in early times, and could not have been, as is, indeed, generally acknowledged, very unhealthy. On the other hand, if the drainage scheme carried out by the kings improved the health of the city, this improvement did not last, for the Rome of the early Empire was so malarious that it was considered very dangerous to remain in it during the autumn months. It may be concluded that Italy did not suffer much from malaria before 200 B.C., that after that date it gradually became more common, as is proved by references to it in literature, until, during the early Empire, many country districts, and also Rome itself, suffered most severely from the disease in an endemic form. The evidence that Rome was highly malarious at the beginning of the Christian era is overwhelming; the more I study the question, the plainer it becomes that the disease exerted a marked influence upon the life of every inhabitant. The poet Horace refers to malaria at least six times, and there are five references in Martial, including three to the deadly

semi-tertian. Horace speaks in the most casual manner of the foolishness of holding out against the disease until the trembling-fit causes disaster at the dinner-table. All who could do so left Rome in the summer, but the poorer people must have suffered severely, as they appear to have slept very often in booths, or open places, thus offering themselves as easy victims to the mosquito. Dropsy, a frequent result of malaria, seems to have been extremely common. Horace says that a man who will not take exercise will certainly fall a victim to it, just as we might say that a sedentary occupation must cause liver-trouble.

Here I should like to meet a possible objection. If malaria was introduced late in the history of Greece and Rome, why have we no mention in ancient writers of the time when it first made its appearance? But it must be remembered that on its first introduction malaria would certainly be confused with other fevers (typhoid, for instance) already existing in the country. In fact even now some kinds of malaria are so like typhoid that the microscope alone can distinguish between them. It would be only after some time that malaria could be recognised as a separate disease, and as late as Galen there is much confusion between the remittent forms and other pernicious fevers. But in the case of Italy there is very likely a reference in the historian Livy to the time when malaria first became widely spread over the country. He says that in the year 208 B.C., an epidemic occurred which did not result in many deaths, but caused much lingering sickness. This looks like an epidemic of malaria, and it should be noticed that the date is within the period of the Hannibalic War, when the land was laid waste and favourable conditions were given to the mosquito. On other grounds also, mentioned in this paper, it seems likely that malaria became common about 200 B.C.

The malarial fevers of Greece and Italy were not confined to the regular types. Besides the malignant forms included under the heading "semi-tertian," many other dangerous kinds are distinctly mentioned. In two books of the Hippocratic *corpus*, *Prorrhetics* and *Prognostics*, there are frequent references to blackwater fever, the algide, hyperpyrexial, comatose and other cerebral forms. Again and again mention is made of aphasia, loss of memory, deafness, convulsions and amblyopia as symptoms of certain kinds of malarial

attacks. It is difficult to discover how far these forms were common in Rome, owing to the way in which Roman writers on medicine repeated the remarks of their Greek predecessors, but we are distinctly told that the semi-tertian was extremely prevalent.

Both in Greece and in Italy the geographical conditions favour the development of malaria, as is proved by the prevalence of the disease in modern times. But, for the present discussion, attention must be paid to the neighbourhood of Athens and Rome. Near Athens were two streams, the Cephisus and the Ilissus, which, by partially drying up in summer, favoured the rapid growth of the mosquito. Near the Piraeus was a marshy district which must have proved a continual focus of malaria. Besides these natural breeding-places, the cisterns in which the Athenians kept their water seem to have harboured mosquito larvae. Aristotle tells us that they often contained the larvae of an insect which was, in all probability, *Chironomus*. The streets also of the city were very muddy in wet weather, so that puddles in out-of-the-way places were certainly numerous. I may refer the reader to four able articles on malaria in Greece, by Drs. Savas and Cardamatis, which appear in the viiith (1907) volume of *Atti della Società per gli Studi della Malaria*. The geographical conditions are shown to be well adapted to the growth of the mosquito.

At Rome similar conditions prevailed. The inundations of the Tiber flooded a considerable portion of the land near its banks, which were notoriously unhealthy. The streets seem to have been as muddy in wet weather as were those of Athens, while the *atrium* of each Roman house contained a pool of rain-water which collected into the *impluvium* through a hole in the roof, intended no doubt, at least originally, to let out the smoke from the household hearth.

It should be noticed that just at the time when malaria appears to have become endemic in Attica and Italy (420 and 200 B.C.), severe wars laid the country waste, and prevented for many years the proper control of irrigating streams and canals. This would favour the spread of malaria.

INFLUENCE OF MALARIA UPON GREEK AND ROMAN HISTORY

In estimating the effects of malaria upon the history of Greece and Rome several facts must be borne in mind. In the first place the ancients had no quinine; the disease must have run its course without being mitigated by any efficient remedy. It is therefore probable that, in Greece at least, some element of the race was weeded out. This would be the Northern strain to which, in all probability, the Greeks owed their best qualities. Again, the virulent remittent forms of the disease seem to have been particularly common. The poet Martial, whose works make but a moderate-sized volume, mentions the malignant semi-tertian three times. In Hippocrates frequent reference is made to those cerebral forms of malaria which, in the words of Mason, lead to "permanent psychical disturbances." Malarial cachexia, with accompanying derangement of the digestive system, was very common. Furthermore, the extent to which malaria occupies the medical treatises is, to say the least, surprising. By far the greater number of the fever cases in the Hippocratic writings refer to malaria in its intermittent or remittent forms; while in the Latin author Celsus, who flourished about 50 A.D., other kinds of fever are scarcely mentioned at all, so that in his book *febris* is practically equivalent to malaria.

The Greeks themselves seem to have noticed that malaria often produced strange psychological effects. So much is plain from their use of the term μελαγχολία and its cognates, which, in the common speech, denoted that a man was crazy, neurotic or even mad, while they were almost certainly medical terms originally, denoting malarial cachexia, or, sometimes, the epileptic convulsions which are often to be observed during a malarial attack. The problem is made a little complicated by the fact that Greek medical terms rarely coincide exactly with any now in use, a source of confusion against which the historian must be always on his guard. "Melancholy" denoted a good many kinds of bilious conditions; but when it is observed that the Greeks themselves thought that quartans had their origin in "black bile" (μέλαινα χολή), that Galen declares large spleens to be due to excess of the "melancholy" humour, that cases of "melancholy" are said to be common in autumn, it seems practically certain that the word was often used to describe malarial states, and that the Greeks

observed how malaria affects the temper, rendering the patient morose and cross-tempered. It is interesting to note that the word μελαγχολία becomes common in Attic literature just at that time (the last quarter of the fifth century) when it seems likely that malaria first became endemic in Attica. Plato in the *Timaeus* declares that vice is due to bodily disease, and in particular derives peevishness, melancholy, rashness, cowardice, forgetfulness and stupidity from bilious humours finding no outlet from the body.

There is a remarkable reference to the influence of malaria upon character in the Hippocratic treatise *Airs, Waters and Places*. "Those who dwell in hollow, hot districts," says the writer, "where the winds and water are warm, are neither tall nor straight. If they drink the water of the place they have diseases of the spleen and stomach. They are stout and fleshy, dark-coloured and bilious. By nature they are neither courageous nor of great powers of endurance. But there are no ill effects when the water is drained off."

What, then, is the change of character which accompanied the decline of Greece and Rome? Between 450 and 300 B.C. the Greeks (at any rate the Athenians, for it is of them that we know most) lost their manly vigour and intellectual strength. Patriotism was still considered a virtue, but few had the energy and initiative to translate theory into practice. Love of ease and comfort grew apace. Philosophy became pessimistic, and there was much brooding over death. In art there appeared a tendency to sentimentalism. The Greek of the third century B.C. was unequal to the effort for further progress, and never recovered the vital force he once possessed.

In Rome and Italy the change was different. The Roman of the early Empire can scarcely be called weak. But he had changed. His sternness had become brutality. He was no longer contented with a simple life, but loved gorgeous display and magnificent banquets. Furthermore, the population of the city changed. The old Romans, apparently, grew fewer in numbers, for there is a constant lament that families were small, while crowds of foreigners flocked to the imperial city, many of whom rose to power and influence. The armies were often recruited from Spaniards and other more virile stock. Historians and moralists repeated *ad nauseam* the truth that the old Roman spirit was dead.

Now it would be absurd to maintain that all these changes were

produced by malaria. The Greek outgrew his city-state, lost his faith in religion, and exhausted his strength in a series of suicidal civil wars. He practised unnatural vice to an extraordinary extent, and this, with other excesses, produced the natural consequences. But surely these influences must have been aided in their operation by the presence of an insidious foe, which weakened the individual from his birth, and left him an easier victim to the disintegrating forces of his environment.

The Roman also outgrew his institutions, and no longer found satisfaction in political life. The farms, which had bred a strong race of yeomen, gradually gave place to large grazing estates. Corn could be imported from abroad more cheaply than it could be grown at home, and the farmers crowded into the already-congested metropolis. Economic causes, then, as well as political and psychological forces, were at work both in Greece and in Italy during the period of decline. But the fact remains that the Greeks became a race of inefficients, while the Romans of the empire may be roughly divided into two classes—a few luxurious debauchees and a host of debased and poverty-stricken retainers.

It is much to be regretted that scientists have paid but little attention to the effects of malaria upon national prosperity and national character. The economic effects, indeed, are noticed with more or less detail by many observers. Celli in his *Malaria* talks of the loss of labour and production caused by the disease, and Clemow describes the appalling incapacitation and economic loss which accompany its ravages. But its influence upon character has never been thoroughly investigated. North, in his fascinating work *Roman Fever*, does say something on the point, but confines himself to the general statement that a highly malarious district, if left to itself, must contain a population that tends to moral degradation. Professor Nieuwenhuis, of Leyden, who has studied the wild tribes of Borneo more than any other traveller, writes to tell me that in that island malaria actually has the disintegrating effect which I assert it had among the Greeks. The results of his investigations are to be found in his book *Quer durch Borneo*. But there certainly is room for a book containing an adequate study of this question from both the physical and the psychological standpoints. A necessary preliminary to such work seems to be a historical study of the dates

at which malaria was introduced into various districts, and of the effects which followed this introduction. Up to the present our information is very slight, being confined to such instances as the invasion of Mauritius by malaria in 1866.

There is also a pressing need for an investigation into the moral and intellectual characteristics prevalent in highly malarious districts, and a comparison of them with those of neighbouring parts, under similar economic and political conditions, but untroubled by malaria. This is another unworked field for historians. I take it that the effects of malaria are threefold:—

(1) It may kill, or drive away, the inhabitants. There is evidence that the population of Greece gradually declined, and that an inferior race, but one relatively immune to malaria, supplanted in course of time the older population. I hope to publish this evidence shortly.

(2) It may cause physical and mental degeneration by making childhood unhealthy.

(3) It may cause inactivity by punishing over-exertion and fatigue.

The first thing to do will be to collect material, and for some time I have been impressing upon anthropologists the necessity of observing the psychological peculiarities of peoples among whom malaria is endemic. It is probable enough that the lapses into barbarism which so often disgrace Europeans living in tropical countries are at least partially caused by this disease or its *sequelae*. I am told that German officials are forced by the Government to carry quinine with them when they set out for malarious regions of Africa, and the reason assigned is that moral deterioration may follow malaria unchecked by prophylactic measures. Unfortunately, I have been unable to discover from written evidence whether this is correct.

CONCLUSION

It is certain that both Greece and Italy were, at least in their most important centres, highly malarious during the period of their decline. Whether Athens and Rome were malarious when they were growing in power and greatness it is impossible to state for certain, but the disease was in all probability rare, even if it was present at all. On the other hand, the decline was in both cases accompanied

by an increase of malaria. The change in the Greek character was just that which we should expect malaria to produce in a highly sensitive and cultivated people, while the savage brutality of the later Romans may be due to the same cause. The peculiar effects of a disease on national morality will certainly vary with the prominent national characteristics. The more effeminate Greek grew weak and inefficient; the stern Roman became viciously cruel. But it must always be carefully remembered that other factors, physical and psychological, contributed to the change in both cases. The growth of intelligence, resulting in dissatisfaction with existing institutions; the decay of agriculture or trade; the slow effects of vice and luxury; the exhaustion of the conditions which stimulate a people to aspire to national greatness—all these were doubtless important factors in the decline of Greece and Rome. But malaria gave rise to physical conditions which afforded an excellent opportunity for other influences to produce their full effect. It must also be remembered that these conditions would be reproduced from generation to generation, for malaria was continuously present, apparently increasing until nearly everybody was more or less infected. A temporary disaster, whether it be war or a virulent epidemic, often stimulates to great deeds and heroic actions; but a weakening endemic disease, attacking every fresh generation as it is born, gives the nation which is its victim no chance to recuperate.

In the preceding pages I have indicated very briefly the main conclusions which it seems just to draw from a study of the classical literatures. But only the fringe of the subject has been touched, even within the limited area to which I have confined my own investigations, and a vast field remains to be worked by those who will take the trouble to trace the effects of malaria upon other nations. It is only from a comparison of results obtained in a series of enquiries that any really valuable knowledge can be expected. The influence of diseases other than malaria is also most important, and should be carefully studied. Mr. T. Spencer Jerome, who has for many years paid attention to the biological aspect of history, sends at my request the following note on the transformation of the Roman character. The remark it contains about pestilence killing off those of the greatest nerve force is interesting, as it is just possible that the great Athenian plague of 430 B.C. weakened the general health by attack-

ing and killing those best fitted to be parents of vigorous offspring. It is a fact that the Athenian population was permanently reduced by this epidemic, and perhaps malaria, which appears to have become endemic in Attica soon after 430, found a people already weakened and less able to resist its ravages.

NOTE

The influence of diseases on historical development will obviously be mainly through their effects in weakening individual nervous systems, and, as a result of this, individual character. Now it seems well established that all infectious fevers play an important part in the etiology of the psychoses and neuroses. Typhoid especially is incontestably a cause of psychasthenia, and often profoundly alters the whole nervous system. So also influenza. The number of severe infectious maladies prevalent throughout Roman history is well known. After the great outbreak of pestilence in the time of Marcus Aurelius, it kept reappearing from time to time for a century or more.

Another point deserving consideration is this:—Is it true that the neurasthenic person is relatively immune to other and more immediately dangerous diseases? Dr. Beard, whose studies of neurasthenia are so well known, asserted this positively—though the explanation he gives seems slightly fanciful.* Dr. V. C. Vaughan, who examined the matter of the typhoid outbreaks in the American army camps at the time of the Spanish war, informed the writer that the fever seemed to select the soundest men. If this be so, then a long continuance of severe infectious maladies would operate not only to produce much nervous debility, but also by a kind of inverse natural selection to eliminate the relatively sound elements in a people.

Now that the biological method of approaching historical problems has been begun, we may hope for real progress, and may cease to rely for explanation of historical developments on hypostatized virtues, vices, institutions and the like. But, as a preliminary step to the application of biology to Roman history, we need to clarify our knowledge of just what were the changes in the average human character of that period, and the order of their development in time. One may well be sceptical as to the extent and accuracy

* Sexual Neurasthenia, by Dr. G. M. Beard, 4th Ed., pp. 63-4, 72-3, 76-8, 116.

of our comprehension of these facts. It is probable that Tacitus, Juvenal and Pliny, as well as most other writers of imperial times, throw more light upon their own character than upon those depicted by them, while our knowledge of early Roman character is involved in an iridescent haze.

T. S. JEROME

CAPRI, ITALY

PLUTARCH ON HEALTH

Let us see whether the theory put forward in the preceding pages is confirmed by a Greek work which was written about the end of the first century after Christ.

Among the *Moral Treatises* of Plutarch is included a work the Latin name of which is *de tuenda sanitate praecepta.* It is intended for the use of those who devote themselves to study or politics (137 c), and lays down the rules which must be observed by such if they wish to keep in health. At first sight it appears to be a sensible but somewhat commonplace series of remarks; but a more careful reading proves that it throws a flood of light upon the hygienic conditions of the period when it was composed. Before proceeding I will give a short analysis of the contents.

It is necessary to keep the hands warm, as chill in the extremities invites fever (123 A). It is useful to accustom the body when in health to the diet which would be necessary in illness, and it should not be thought insufferable to dine unbathed (123 B—D). The body ought to be nourished, as a rule, with simple foods, so that, should an occasion occur when feasting cannot be avoided, no harm results from indulgence. If some high official invite us, or other imperative call come when we are indisposed, it will be less boorish to abstain than to fall into πλευρῖτις or φρενῖτις through false shame (123 E—124 D).

Food and drink are to satisfy hunger and thirst. Dainties should not be consumed merely because they are costly, or because we wish to boast that we have eaten them. The body must not tyrannise over the soul, nor yet the soul over the body, so as to cause over-indulgence. A man should take a pride in his power to abstain. Rich, tempting dishes cause us to eat too much (124 E—126 B).

Pleasure is impossible without health. We are wont to neglect plain living when we are well, and in sickness to lay the blame upon climate (ἀέρας, χώρας), instead of our own intemperance. When

ill we should say to ourselves that drinking cold water, or an untimely bath, has deprived us of many pleasures. In this way we are made more careful when in health (126 B 127 B).

Granted that fevers are caused by exertion, heat and chill, too much food increases the liability (127 B—D).

The forewarnings given by fever must not be neglected. Some, when they feel an attack coming on, betake themselves to baths and banquets, lest they fall ill before they have satisfied their desires; others, more refined (*κομψότεροι*), are ashamed of showing that they are unwell, and obey the call of their companions; most men hope that the feeling of uneasiness will pass off. But on the morrow they have to confess to catarrh, fever or colic. Then they will beg the doctor to allow them wine or cold water. All such should remember that the unhealthy body feels no pleasure in the indulgences which caused the trouble (127 D 128 E).

The over-strict diet of one who is always afraid of his health giving way is certainly to be blamed, as it renders the body liable to fall sick, while it makes the spirit timid and unenterprising; but it is certainly unwise to wait for those internal pains which are the forerunners of fever before moderating one's desires and appetites. It is necessary also to be on the watch for bad dreams, crossness of temper and melancholy (128 E—129 C).

When visiting a sick friend enquiry should be made whether it was plethora, heat, exertion, lack of sleep or wrong diet that caused his fever. His answers will serve as a guide. One should care for one's own mode of life, avoiding all excess (129 D—130 C).

Reading and discussion are excellent physical training. The mockery of inn-keepers or muleteers can be neglected (130 C 131 B).

After exercise, cold baths are to be avoided. Those who so indulge fall ill, unless they follow in the smallest details that strict diet which is so undesirable. It is better to oil the body near a fire (131 B—D).

Meat, dried figs and cooked eggs are not desirable; vegetables, fowl and light fish are to be the staple food. Milk as a drink should be avoided, wine in moderation is good, but not as a "pick-me-up" after exposure; water should be drunk several times a day. If it be thought a shame to be deprived of food when a fever is imminent, water may be drunk (131 D 132 F).

While eating, a man should exercise his mind with a book or

conversation. This will make him less attracted by the pleasures of the table (133 A—134 A).

Emetics and purges are bad. Dieting is the proper remedy for indigestion. If something must be done, vomiting is the less evil, but violent drugs must be avoided. Drinking water or fasting for a few days may be tried, or even an injection. Most people take refuge at once in strong purgatives, and suffer for it (134 A—F).

On the other hand, a rigid system of fasting is bad. It is absurd to keep well by ceasing to perform the functions of living. Nay, idleness is not healthy (135 A—136 A).

Toil should not be varied by exhausting pleasures. Love of honourable pursuits will drown any desire that is felt for the latter (136 A—E).

A man should learn all he can about his own constitution, what suits it and what does not. It is important that care be taken not to tax it at the change of the seasons (136 E—137 B).

Students must not tax their bodies by too much study, as the many do by worry and exertion at harvest-time. Otherwise they will be compelled to lay aside their books, while they are recovering from a fever (137 C—E).

It will, I think, be admitted that at the period when the treatise was composed there was much ill-health. The precepts given by the writer himself are strict, and he distinctly states that there were some who imposed upon themselves such rigid rules of life that health was obtained at far too high a cost; for they could not use it without interfering with those prescribed habits which kept them well.* The writer does not seem to be referring to infectious sickness, for he nowhere mentions either contagion or infection.† Indeed, either ancient Greece was singularly free from infectious maladies (other than occasional epidemics) or else the Greeks did not think the danger worth considering. At any rate isolation of the sick, and similar prophylactic measures, were not generally recognised.‡ The great danger, according to Plutarch, was "fever."§ The symptoms of

* 128 E and 131 C.

† When calling on a sick friend the visitor is not supposed to take any precautions (129 D).

‡ Fumigation by sulphur was known as early as the Homeric period (*Odyssey*, xxii, 481, 493), but the medical writers appear to know nothing of it. Did they regard it as a superstition? Certain skin and eye diseases, with consumption, were regarded as infectious, but not fevers (Aristotle, *Prob.* vii, 8.)

§ Mentioned several times—123 A; 127 B, E; 128 A, F; 129 D; 132 E; 137 D. Once a more specific name, φρενῖτις, is given (124 B).

fever are not described, but a warning is given not to neglect the premonitory signs, and among these are crossness of temper and melancholy.* But the risk of falling ill of fever is said to be greatly increased by certain actions or habits. The "causes" of fever include:—

(1) Violent fatigue (*κόπος*).†

(2) Extremes of temperature,‡ especially chilled extremities and cold baths at unseasonable times.§

(3) Over-indulgence in food and drink.‖

(4) Insufficiency of rest and sleep.¶

In addition to these definite dangers, the general tone of the treatise implies a strong recommendation to avoid taxing the body or mind by excess in any form.

Now, with the exception of that prophylaxis which is the direct result of modern discoveries,** this advice is just that which is now given to those who dwell in malarious regions.†† If it be urged that it is unsafe to conclude that malaria is the disease which our writer had in mind, inasmuch as moderation is a good rule under any climatic conditions, the following points should be considered.

Only in a highly malarious country can it be said that fatigue will bring on fever. Of course, fatigue may be the indirect cause of a feverish chill, but it is in malarious countries that the necessity of avoiding over-exertion, and the practical certainty of an attack should this rule be violated, become painfully obvious from repeated experiences. It is also remarkable that such stress is laid on keeping the hands warm. In a non-malarious country cold in the extremities can scarcely be considered a grave danger, but in a malarious region great care must be taken to avoid chill. It should also be noticed that the change of the seasons is regarded as an

* 129 C.

† 127 B, E; 129 D.

‡ 127 B (*δι' ἐγκαύσεις καὶ διὰ περιψύξεως*); 129 D.

§ 123 A; 126 F; 127 E; Cf. also 131 C.

‖ 124 B; 127 C, E; 128 A; 129 D; Cf. also 137 C.

¶ 129 D (*ἀγρυπνίαν*); 137 D.

** E.g., the use of quinine and mosquito-netting.

†† See Celli *Malaria* and Manson *Tropical Diseases* (malaria).

especially dangerous period,* and malaria is most common just before the autumnal equinox. Particular attention should be paid to the difficult passage 137 C,† the general drift of which seems to be that poor country-folk constantly fall ill during their exertions at harvest-time. Indigestion and constipation were evidently common complaints when the writer lived, as he tells us how the people took refuge in violent purgatives.‡ Now although there are many causes of these stomach complaints, derangement of the digestive organs is the invariable accompaniment of malarial cachexia. If any doubt still remains as to the kind of fever which is referred to, the use of *φρενῖτις* § in 124 B should dispel it at once. This word is certainly employed by the medical writers to denote a very pernicious kind of remittent malaria.‖

I have tried to prove in the above discussion that the Greeks of the first century A.D., or at any rate some of them, were liable to malarial attacks whenever they put the body to unusual exertions; in other words that the country, or some part of it, was highly malarious. What must have been the consequence to the people at large? What nation can prosper or develop if strain has to be avoided at all costs? How are war, commerce and successful agriculture to be carried on under such conditions? Surely nothing but stagnation is possible. Now the literary evidence shows that malaria was common in Greece during the fifth century, since tertian and quartan fevers are constantly referred to in the Hippocratic writings. But there are only two allusions before 500 B.C. which can point to malaria (*πυρετὸς*¶ in *Iliad* xxii, 31, and *ἠπίαλος* in Theognis 174), and both of these are doubtful. Hesiod never mentions fever among

* 137 B.

† *ὅσα μὲν γὰρ μικρολογίας καὶ ἀνελευθερίας προσκρούσματα λαμβάνουσιν οἱ πολλοὶ περί τε συγκομιδὰς καρπῶν καὶ τηρήσεις ἐπιπόνους, ἀγρυπνίαις καὶ περιδρομαῖς ἐξελέγχοντες τὰ σαθρὰ καὶ ὕπουλα τοῦ σώματος, οὐκ ἄξιόν ἐστι δεδιέναι μὴ πάθωσιν ἄνδρες φιλόλογοι καὶ πολιτικοί.*

‡ *φαρμακείας, ἐφ' ἃς οἱ πολλοὶ φέρονται προχείρως* (134 F).

§ For *φρενῖτις* see Hippocrates, Kühn II, 27, 28, 299, 300, 387.

‖ See Littré's *Hippocrates*, Vol. II.

¶ The word *πυρετὸς* does not occur again till the second half of the fifth century B.C. In Homer it was said by some ancient commentators to mean "heat." Why did they not assume the meaning to be fever? Surely this points to a tradition that malaria did not exist in Homer's time.

the plagues of the Boeotian farmer, but Plutarch, a native of Boeotia, has a different tale to tell. There can, I think, be no doubt that malaria was rapidly on the increase§ from the fifth century B.C., and that it was largely responsible for the lack of energy that the Greeks began to exhibit during the fourth century.

Plutarch noticed that the feverish attack is often preceded by melancholy and crossness of temper,* two most noticeable symptoms of malaria. The physician Hippocrates went much further. After carefully describing what regions are most malarious, he goes on to say that the inhabitants of such regions are stunted in growth, and have neither courage nor powers of endurance.† Plato declared that ill-health (the symptoms point to malaria as much as anything else) produces crossness, melancholy, rashness, cowardice, forgetfulness and stupidity.‡ Surely it is reasonable to conclude, not only that malaria produced disastrous changes in the Greek character, but also that the great thinkers among the Greeks were perfectly conscious that it did. Plato would have introduced remedial measures,‖ but, of course, his advice was not taken, and Greece gradually fell into the unhealthy state which is manifest in the pages of the treatise *de sanitate tuenda*.

§ The words πυρετός, πυρέσσω, become more common, and are regularly used in non-medical works to mean intermittent (i.e. malarial) fevers. See Demosthenes, 118, 20.

* 129 C.

† *Airs, Waters, Places*, Kühn I, 566, 567.

‡ *Timaeus* 87 A.

‖ Namely, careful nurture and good institutions (*Timaeus* 87 B.). This reminds one of the remark of Hippocrates (*loc. cit.*) that νόμος might restore the moral fibre of malarious peoples, and good drainage bring back their health. For Plato's later views on vice see Gomperz, *Greek Thinkers*, Vol. III, 225, 226.

TWO NEW HUMAN CESTODES AND A NEW LINGUATULID

TWO NEW HUMAN CESTODES AND A NEW LINGUATULID

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I. A NEW BOTHRIOCEPHALID IN MAN

(*Dibothriocephalus parvus* n. sp.)

Three pieces of a tape-worm, none of which had a head, were received by me from Dr. Elkington, Tasmania, in 1906, with the following history: 'It came from a Syrian, not long arrived from his native country, and aged 37. It was caught on December 20th, 1898, at Launceston, with the aid of *Filix mas*. A quantity had already come away before he came under treatment. So it is possibly a Levantine product after all, and not Australian in origin. He is supposed to have come from Beyrout, but this is not certain.'

A superficial examination shewed that the tapeworm had all the appearance of a *Dibothriocephalus*. The proglottids had a clearly defined central uterine rosette, and with a lens the openings of the cirrus and uterus could be seen, and in several segments the cirrus was extruded. The surface of many of the segments was much corrugated with transverse and longitudinal furrows (probably due partly to the preservative and partly to erosion (digestion) of the surface.

The strobila consisted of three portions, the lengths of which were respectively 1025, 940 and 690 mm. In the portion, 1025 mm. long, there were 420 proglottids, usually broader than long. The smallest anterior proglottids measure 1·3 mm. broad by 0·6 mm. long. The largest posterior proglottids measure 5·0 mm. broad by 3·0 mm. long. The terminal segments tend to become quadrate and measure 4·0 by 4·0. Several segments approximately quadrate are, however, interpolated irregularly so that the gradual decrease in breadth and

increase in length from one end of the worm to the other is broken at intervals and is not uniform as in *D. latus*, &c. As stated above the dimensions of the largest gravid segments are 5 by 3 mm. The uterus forms a central rosette with four to five loops on each side of the median line, occupying about the middle half of the length.

In a proglottid measuring 3·5 by 2·25 the genital atrium is situated 0·4-0·5 mm. behind the anterior margin, and the uterine opening is situated the same distance behind the genital atrium.

The eggs, operculated, average 59·2 by 40·7 μ. Calcareous corpuscles were absent in those segments examined.

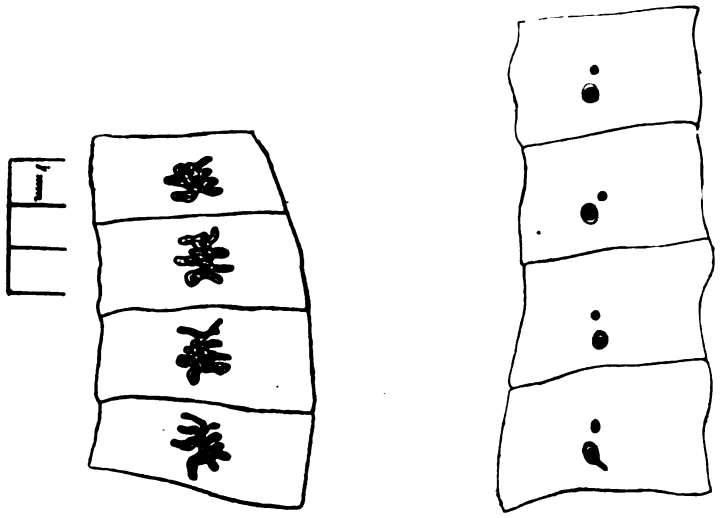


FIG. 1. *Dibothriocephalus parvus*

(1) Outline of uterine rosette
(2) Genital atrium, uterine pore and protruded cirrus

The question now arises as to the identity of this worm.

(*a*) On comparing it with *D. latus* we see that the maximum width of gravid segments is 5 mm., while the minimum width of gravid segments of *D. latus* is 10-12 mm., and the maximum width 20 mm., so that the worm is a very much smaller one, as is evident to the naked eye. This is also shewn by the size of the quadrate segments which in *D. latus* are 6 by 6 mm., while in *D. parvus* they are 4 by 4 mm., and moreover the quadrate segments in *D. latus*

occur posterior to segments which attain a width of 15 mm., while in *D. parvus* they occur after segments, the maximum width of which is only 8 mm., and in the case of the interpolated segments, after segments which are still narrower.

(*b*) The whole strobila appears always to be distinctly thicker than *D. latus*, a point which is especially noticeable along the lateral margins.

(*c*) Secondly there is no indication of the serration due to the projection of the posterior lateral angles, a feature well marked in *D. latus*.

(*d*) The eggs of *D. parvus* (59·2 by 40·7 μ) are smaller and rounder than those of *D. latus* (68-71 by 44-45 μ).

(*e*) Calcareous bodies are absent in *D. parvus*, while they are present (few) in *D. latus*.

(*a*) On comparing with *D. cordatus*, we find that this has only 50 immature segments while in one of the pieces of *D. parvus* there were at least 200 segments before maturity.

(*b*) The mature segments of *D. cordatus* are 7-8 mm., the maximum width of *D. parvus* is 5 mm.

(*c*) The quadrate segments of *D. cordatus* measure 5-6 mm. square. Those of *D. parvus* 4 by 4 mm.

(*d*) The uterine loops of *D. cordatus* are 6-8. Those of *D. parvus* 4-5.

(*e*) Eggs. *D. cordatus* 75-80 by 50. *D. parvus* 59·2 by 40·7 μ.

(*f*) Calcareous bodies. *D. cordatus*, 28-30, numerous. *D. parvus*, absent.

The characters of this bothriocephalid seem to me to be sufficiently distinct to warrant the making of it a new species. I propose the name *Dibothriocephalus parvus*. The type species is deposited in the museum of the Liverpool School of Tropical Medicine.

II. A NEW HUMAN CESTODE

(*Taenia bremneri*, n. sp)

In July, 1907, I received from Dr. Bremner, Nafada, Northern Nigeria, about half a dozen segments of a tape worm passed by a Fullani woman. The patient stated "that *all* Fullani women have them, and that they are got through drinking sour milk."

The most striking feature of the proglottids was their size, being greater in length, and especially in breadth, than that of any human *Taenia* so far described. The measurements were as follows:—

Maximum, 32 × 9 mm.
Average, 28·6 × 8·5 mm.
Commonest, 29 × 10 mm.
Smallest, 21 × 6 mm.

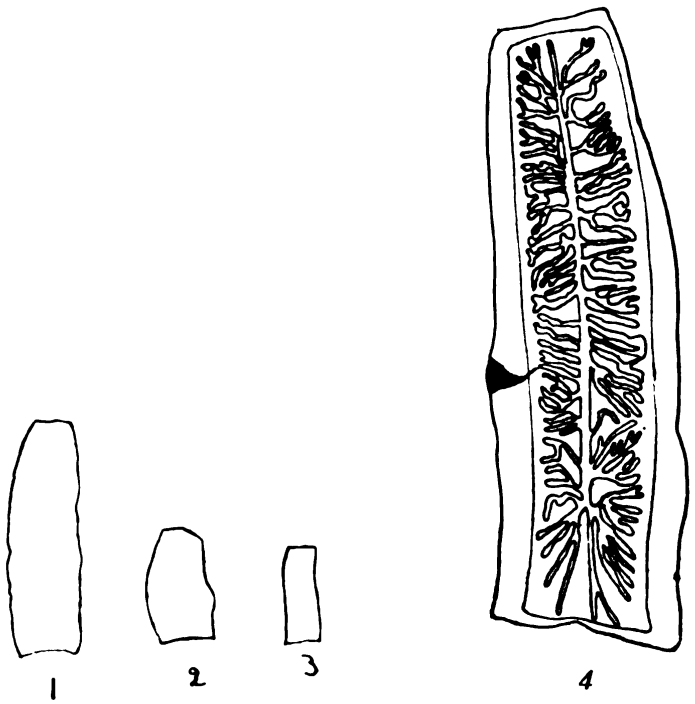


FIG. 2. *Taenia bremneri, n. sp.*

Gravid segments of (1) *T. bremneri*, (2) *T. saginata*, (3) *T. solium*, (4) Uterine branches enlarged nearly four times

On clearing a specimen, the uterine segments were in some segments 22 to 24 in number, not counting the "terminal" one which curves forward and has four or five branches on it. The figure shows the arrangement of the uterus. The genital pore is prominent and lies behind the middle of the segment.

Eggs: Minimum, 34·2 × 30·4.
Commonest, 38 × 30·4.
Maximum, 45·6 × 41·8.

Calcareous bodies are numerous, measuring 15·2μ in diameter. This species is distinguished from some of the species found in man by the following points:—

T. africana.—Segments always broader than long.

T. confusa.—The gravid segments may be as long as 35 mm., but the width is only 5 mm., whereas in the present species, with a length of 32 mm. the width is 9 mm.

T. saginata.—Gravid segments, 15 to 20, rarely 25 mm. long, 4 to 7 mm. wide.

T. solium.—Gravid segments, 10-12 long by 5 mm. broad.

I hope soon to be in a position to describe complete specimens of this worm; for the present I propose the name *Taenia bremneri.*

The type segments are in the museum of the Liverpool School of Tropical Medicine.

III. A NEW LINGUATULID

(*Porocephalus pattoni, n. sp.*)

Habitat:—This linguatulid occurs in the lungs of the Indian rat snake *Zamenis* v. *Ptyas mucosus* or Dhaman.

Diagnosis:—Body greyish white (spirit specimens), showing a separation into head, neck and body. The head is globular and flattened ventrally and rounded dorsally, and is separated from the body by a fairly distinct narrow portion or neck. Of some dozen

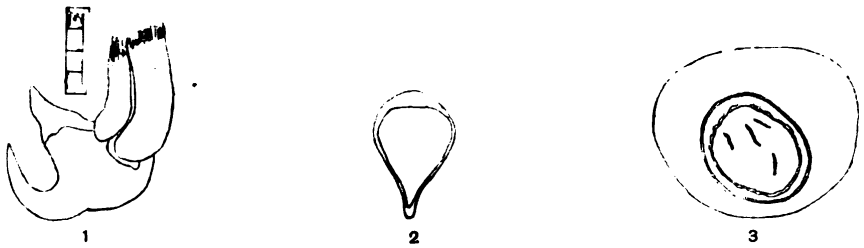


FIG. 3. *Porocephalus pattoni*
(1) Hook, (2) mouth, (3) egg

specimens measured the longest was 11·5 centimetres, while the shortest was 2·5 centimetres. The majority were between 6 and 8 centimetres. The number of rings varies somewhat, but an average size specimen has 36. There are generally two rings on the head. The body is cylindrical, ends bluntly, and the posterior end exhibits some tortion. The thickness of the body is about 2·5 mm. On the

ventral surface of the head are two pairs of strongly curved hooks similar in appearance to those in many other Linguatulids, e.g. *Porocephalus annulatus*, and the details also of the hooks resemble those of other Linguatulids as far as can be judged from figures, though no measurements usually accompany these. In fig. 3 is seen an isolated hook with its appendages. They bear a general resemblance, for instance, to those of *P. teretiusculus*. Between the hooks lies the chitinous opening of the mouth. This has the shape of a horse-collar, resembling the mouth of *P. annulatus*. In front of the hooks and mouth are several small papillae corresponding in position to the continuation of the clefts that contain the hooks. I was unable to detect papillae in any other situation. The cutaneous pores are abundant on the head and anterior rings; they then become indistinct, but some are visible on the last segments. The eggs taken out of the uterus measure about $80 \times 70\ \mu$ (fig. 3).

As far as I am able to judge from the literature of the subject this species is a new one. I propose provisionally the name *Porocephalus pattoni* in honour of Capt. Patton, I.M.S., who was kind enough to send me the specimens.

The type specimens are in the museum of the Liverpool School of Tropical Medicine.

Addenda et Corrigenda

Page 43. *Chrysops dimidiatus*, v. d. Wulp.

The specimens recorded from Banana* are of this species. Those from the other localities are all *Chrysops silacea*, Austen (Ann. and Mag. Nat. Hist., ser. 7, Vol. XX, p. 509, 1907). On comparing the examples of *Chrysops* from the Congo Free State with Austen's description of *C. silacea* I came to the conclusion that we had examples of both *C. dimidiatus* and *C. silacea*. Mr. Austen, who has kindly examined a specimen of the former, says that I am "perfectly right" in my identification; the footnote by Mr. Austen (l. c., p. 512) should, therefore, be amended so as to include only those specimens which were taken by Dr. Ettiene at Banana.

ROBT. NEWSTEAD.

* The package containing these specimens, was dated 'Oct. 22, 1902.'

EXPLANATION OF PLATE L

Fig. 1.—*Dibothriocephalus parvus, n. sp.* Portion of strobila, actual size.

Fig. 2.—*Porocephalus pattoni, n. sp.* The Linguatulids *in situ* in the lung of *Zamenis mucosus*.

Fig. 3.—*Porocephalus pattoni, n. sp.* Actual size.



Fig. 1



Fig. 3



Fig. 2

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